AN EVALUATION OF THE CORTICOTROPIN-RELEASING HORMONE AND LEPTIN GENE SNPS RELATIVE TO CATTLE BEHAVIOUR

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By Kaley Adeene Pugh

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ABSTRACT

Temperament in cattle, defined as an animal's response to handling by humans, had been associated with production traits such as average daily gain and meat quality, and can also be considered a welfare issue. Temperament is a stress response trait, and therefore the hypothalamic-pituitary adrenal (HPA) axis likely plays a role in determining individual animal's responses. The purpose of this study was to examine whether there are associations between single nucleotide polymorphisms in two genes involved in both the HPA axis and growth, Corticotropin-releasing hormone (*CRH*) and Leptin (*LEP*), and various measurements of temperament in beef cattle. In this study, 400 crossbred beef steers were evaluated over three sessions using a traditional subjective score and three objective measurements of response to handling: Strain Gauge (absolute strain force, ASF), Movement Measurement Device and Exit Time (ET) as well as habituation for all measurements (session 3 values – session 1 values). Backgrounding growth and final carcass data were also collected. The steers were genotyped at three previously reported SNPs: *CRH 22C>G*, *CRH 240C>G* and *LEP 73C>T* by PCR-RFLP. Subsequently, the genotypes and two-way interactions between *LEP* and each *CRH* SNP were analyzed as effects on the various temperament, growth and carcass measurements. There was a significant interaction between *LEP* and *CRH 240C>G* for ASF 1, ET 3 and ET 3-1, with the *LEP CC/CRH 240C>G CC* genotype appearing favorable. Additionally, the *LEP CC/CRH 22C>G GG* genotype appears to be favorable for ASF 1. These results indicate that it may be possible for cattle producers to select for favorable temperament using these SNPs, however these results should first be validated in additional populations.

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TABLE OF CONTENTS

PERMISSION TO USE I
ABSTRACT II
ACKNOWLEDGEMENTS III
TABLE OF CONTENTS IV
TABLE OF TABLES VI
TABLE OF FIGURES VII
LIST OF APPENDICES VIII
LIST OF ABBREVIATIONS IX
1 INTRODUCTION 1
2 LITERATURE REVIEW 2
2.1 Behaviour 2
2.1.1 Fear/Anxiety, Temperament & Personality 2
2.1.2 Measurement of fear/anxiety 3
2.1.2.1 Rodents 3
2.1.2.2 Cattle and other livestock 4
2.1.3 Associations with production 6
2.1.3.1 Other livestock 6
2.1.3.2 Cattle 8
2.1.4 Genetics of fear/anxiety response 10
2.1.4.1 Rodents 10
2.1.4.2 Cattle & Other livestock 10
2.1.5 Physiology of fear/anxiety response 12
2.1.5.1 The HPA Axis 12
2.2 CRH 16
2.2.1 *CRH* Gene structure and location 16
2.2.2 *CRH* Gene expression 19
2.2.3 Effects of CRH 19
2.2.4 Mutations in *CRH* 20
2.2.4.1 Mutations in cattle 21
2.3 LEP 21
2.3.1 *LEP* Gene structure and location 21
2.3.2 *LEP* Gene expression 22
2.3.3 Physiological effects of LEP 22
2.3.4 Mutations in *LEP* 23
2.3.4.1 Mutation in cattle 23
2.4 Objective & Hypothesis 24

3 MATERIALS & METHODS 25
- 3.1 Animals & Processing 25
- 3.2 Behaviour measurements 27
  - 3.2.1 Subjective Score 28
  - 3.2.2 Strain Gauge 29
  - 3.2.3 Movement Measurement Device 32
  - 3.2.4 Exit Time 33
  - 3.2.5 Habituation 34
- 3.3 Blood Collection & DNA extraction 34
- 3.4 PCR-RFLP 34
  - 3.4.1 *Leptin 73C>T* 34
  - *3.4.2 CRH 22C>G* 35
  - *3.4.3 CRH 240C>G* 36
- 3.5 Growth & Carcass measurements 36
  - 3.5.1 Backgrounding 36
  - 3.5.2 Finishing & Carcass Data 37
- 3.6 Statistical analysis 37

4 RESULTS 39
- 4.1 PCR-RFLP 39
- 4.2 Descriptive Statistics 42
  - 4.2.1 Behaviour Measurements 42
  - 4.2.2 Habituation 44
  - 4.2.3 Growth & Carcass measurements 48
- 4.3 Statistical Analysis 50
  - 4.3.1 Simple Regressions 50
  - 4.3.2 Mixed Model ANOVA: Behaviour and genotype 54
  - 4.3.3 Mixed model ANOVA: Carcass measurements & genotype 62

5 DISCUSSION 63
- 5.1 Allele Frequencies 63
- 5.2 Behaviour Measurements & Habituation 63
- 5.3 Associations of Genotype with Growth and Carcass Measurements 66
- 5.4 Behaviour & Genotype 66

6 CONCLUSIONS 69

7 LITERATURE CITED 70

APPENDICES 80

TABLE OF TABLES

Table 3.1 A timeline for all procedures performed on the cattle....26
Table 3.2 The 1-5 scale used for allocation of a Subjective Score....28
Table 4.1 Number of steers of each SNP genotype and allele frequencies for each SNP....41
Table 4.2 Summary of the behaviour measurements from the first and third measurement sessions....43
Table 4.3 Tests for normality results for the behaviour measurements....43
Table 4.4 Summary of habituation data for the behaviour measurements....45
Table 4.5 Paired t-test results for the differences between means for each measurement between sessions 1 and 3....45
Table 4.6 Summary of growth and carcass measurements after backgrounding....48
Table 4.7 Summary of growth and carcass measurements after finishing....49
Table 4.8 The number (and %) of carcasses in each Quality Grade....49
Table 4.9 Results of the simple regression analysis between the number of LEP T alleles and the behaviour measurements....50
Table 4.10 Results of the simple regression analysis between the number of *CRH 22C>G* G alleles and the behaviour measurements....51
Table 4.11 Results of the simple regression analysis between the number of *CRH 240C>G* G alleles and the behaviour measurements....51
Table 4.12 Results of the simple regression analysis between the number of *LEP* T alleles and the growth and carcass measurements....52
Table 4.13 Results of the simple regression analysis between the number of *CRH 22C>G* G alleles and the growth and carcass measurements....53
Table 4.14 Results of the simple regression analysis between the number of *CRH 240C>G* G alleles and the growth and carcass measurements....54
Table 4.15 A summary of the preliminary Tests of Main Effects for SNP genotypes on behaviour....55
Table 4.16 LSM for behaviour measurements where there was a significant effect of *CRH 240C>G* genotype....56
Table 4.17 LSM for traits where the interaction between *LEP* & *CRH 240C>G* genotype was significant....57
Table 4.18 LSM for the trait where the interaction between *LEP* & *CRH 22C>G* genotype was significant....57
Table 4.19 A summary of the tests of main effects for SNP genotypes on growth and carcass traits....62

TABLE OF FIGURES

Figure 2.1 The hypothalamic-pituitary adrenal axis stress response pathway 14
Figure 2.2 Multiple sequence alignment for CRH... 18
Figure 3.1 A schematic representation of the temperament measurement sequence 27
Figure 3.2 A picture of the head gate to which the strain gauge device was attached 30
Figure 3.3 A picture of the strain gauges in place on the bar of the head gate, shown with a quarter for scale.. 30
Figure 3.4 A graphical representation of the raw Strain Gauge data from two steers.. 31
Figure 3.5 The metal panels used to form the Exit Time test chute 33
Figure 4.1 A representative gel for the *LEP* PCR-RFLP 39
Figure 4.2 A representative gel for the *CRH 22C>G* PCR-RFL............................. 40
Figure 4.3 A representative gel for the *CRH 240C>G* PCR-RFLP 40
Figure 4.4 Average Subjective Score for each measurement session....................... 46
Figure 4.5 Average Absolute Strain Force for each measurement session............... 46
Figure 4.6 Average number of MMD Peaks for each measurement session 47
Figure 4.7 Average Exit Time for each measurement session 47
Figure 4.8 The interaction between *LEP* & *CRH 240C>G* genotypes for Absolute Strain Force 1 58
Figure 4.9 Interaction between *LEP* & *CRH 240C>G* genotypes for Exit Time 3.. 59
Figure 4.10 Interaction between *LEP* & *CRH 240C>G* genotypes for Exit Time 3-1.. 60
Figure 4.11 The interaction between *LEP* & *CRH 22C>G* genotypes for AbsoluteStrain Force 1 61

LIST OF APPENDICES

APPENDICES 80

Appendix A Backgrounding Diet 80
Appendix B Finishing Diet 81
Appendix C Steer Genotypes at LEP, CRH 22C>G & CRH 240C>G SNPs 82
Appendix D Behaviour Measurements 87
Appendix E Backgrounding Growth Data 100
Appendix F Finishing Growth & Carcass Data 109
Appendix G Abstract 122
Appendix H Poster 123

LIST OF ABBREVIATIONS

μl	microliter
ACTH	adrenocorticotropin
ADG	average daily gain
ASF	absolute strain force: sum of the absolute values of the strain gauge readings, used to compare sg data between animals
Avg.	average
CRH	corticotropin-releasing hormone
ddH_2O	double distilled water
DMSO	dimethyl sulfoxide
DNA	deoxyribonucleic acid
dNTPs	deoxynucleotide triphosphates
EDTA	ethylene diaminetetra acetic acid
EOB BF	end of backgrounding back fat, measured by ultrasound
EOB REA	end of backgrounding rib-eye area, measured by ultrasound
EPD	expected progeny difference
ET	exit time
HPA	hypothalamic-pituitary adrenal
JB	jeffrey's buffer
Kb	kilobase
LEP	leptin
LSM	least squared mean
$MgCl_2$	magnesium chloride
mM	millimolar
MMD	movement measurement device
mV	millivolts
Ng	nanogram
PCR	polymerase chain reaction
Pmol	picomole
POMC	pro-opiomelanocortin
REA	rib-eye area
RFLP	restriction fragment length polymorphism
RIA	radioimmunoassay
SD	standard deviation
SEM	standard error of the mean
SG	strain gauge
SNP(s)	single nucleotide polymorphism(s)
SOT Wt	start of test weight
SS	subjective score
TE	tris – edta
WC Wt	warm carcass weight

1 INTRODUCTION

Profitable beef production requires a combination of various factors, including proper nutrition for the type of cattle and target market, effective herd health management and efficient reproduction, among many others. Another important aspect of cattle management, that is sometimes overlooked, is the temperament of the livestock themselves. For the purposes of this thesis temperament will be defined simply as an animal's response to handling by humans, in accordance with Burrow (1997).

There are several reasons to study, and select for, temperament in cattle. Perhaps the most intuitive reason is that cattle with calmer temperaments are easier to handle and cause fewer injuries to themselves, their handlers and other animals. While this statement has yet to be empirically tested, it is interesting to note that Schmutz *et al.* (2001) discussed how two heifers with excitable temperaments broke their jaws in one night, and in this study a highly agitated steer broke its hip during one of the weighing sessions. These incidences illustrate the types of serious injuries to animals that could potentially be prevented by conscientiously selecting for good temperaments in breeding stock.

Several studies have also shown that cattle with calmer temperaments have better ADG (average daily gain) (Voisinet *et al.* 1997; Fell *et al*. 1999), as well as better meat quality (Voisinet *et al.* 1997b; King *et al.* 2006; Kadel *et al.* 2006). Given these associations, there is further economic impetus for producers to include temperament in their selection criteria.

2 LITERATURE REVIEW

2.1 Behaviour

Behaviour can be defined as an organism's response to a stimulus, therefore the general study of animal behaviour encompasses a broad variety of topics including learning, parenting, feeding and predator response behaviours, among innumerable others. The specific aspect of animal behaviour research that will be addressed in this literature review is animals' fear/anxiety responses to aspects of their physical environment, including the response to handling by humans.

2.1.1 Fear/Anxiety, Temperament & Personality

Animals' reactions to stimuli are often grouped into specific and distinct types of responses such as fear (Boissy 1995) and anxiety (Ramos *et al.* 1997) or traits such as temperament (Burrow 1997) and personality (Forkman *et al.* 1995). Fear and anxiety are responses to threats, where fear is generally considered to be the response to a real threat, while anxiety is a response to a perceived or potential threat (Boissy 1995).

Temperament and personality are broad, individually consistent traits that describe an animal's general response in fear or anxiety producing situations. For the purpose of this review and the experiment described later, when temperament in domestic livestock is discussed it will be according to the specific definition given by Burrow (1997), that temperament is an animal's reaction to handling by humans.

2.1.2 Measurement of fear/anxiety

The methods used for measurement of fear/anxiety behaviours in livestock are generally derived from techniques used in rodents, therefore some of the behaviour measurement techniques used in rodents will be briefly discussed. This will be followed by a short overview of the different techniques used to measure temperament in cattle. The measurements used in other livestock such as sheep, swine and poultry will also be briefly discussed as much of the interesting work performed regarding the genetics, physiology and production effects of fear/anxiety has been conducted in these animals.

2.1.2.1 Rodents

Extensive studies have been performed with rodents in regards to fear and anxiety related behaviours. According to Boissy (1995), the classic fear test used in rodents – the open field test – was developed by C.S. Hall in 1934, and is widely used because it involves several stressful elements including a novel environment, lack of shelter, and bright light. The modern open field test is fairly consistent between investigators, and generally follows a set up similar to that described by Gershenfeld *et al.* (1997): a square test area, illuminated from overhead, is divided into equal sections by a photocell sensor system; an animal naive to the apparatus is placed inside, and measurements such as total distance traveled, horizontal activity, number of times the animal rears up, time in the center of the field, and percentage of time active or resting are measured. The number of defecations and/or urinations are also occasionally measured (Ramos *et al.* 1997). The open field test has been adapted for use as a measure of fear/anxiety in a variety of other species including chickens, cattle, pigs, sheep (as reviewed by Boissy 1995), and even animals such a foxes (Harri *et al.* 1995). In the open field test, increased time in the center of the field (considered aversive) and increased locomotion are taken as indicators of low fearfulness (Ramos *et al.* 1997).

Another commonly used fear/anxiety test in rodents is the elevated plus maze. Like the open field test, the elevated plus maze apparatus is generally consistent between investigators, following a set up similar to that described by Ramos *et al.* (1997). Their apparatus consisted of four arms (45x10 cm) arranged in a "+" and a central intersection area (10x10 cm), all of which was 66 cm above the ground; two of the arms were enclosed by 50 cm walls, while the other two had no walls. Ramos *et al.* (1997) recorded the number of entries into each arm, the time spent in each arm, and the number of partial entries followed by retreats into each section – these measures are typical of those recorded by other investigators using this apparatus. In the elevated plus maze test the open arms are considered aversive, and thus increased time in the open arms is considered to indicate low fearfulness (Ramos *et al.* 1997).

Other common tests of fear/anxiety used in rodents include forced swimming and water maze tests (Crabbe *et al.* 1999), the light/dark box test (Ramos *et al.* 1997), response to tail suspension (Turri *et al.* 2001), response to novel objects (Radcliffe & Erwin 1998) and acoustic startle response (Fernández-Terul *et al.* 2002).

2.1.2.2 Cattle and other livestock

Temperament in livestock has been studied in a variety of ways. The classic temperament measurement is the subjective score. In one of the original cattle temperament studies Tulloh (1961) subjectively rated the behaviour of 72 animals as they entered a scale, a crush, and a bail (headgate) as well as their behaviour while restrained in the bail. The behaviour in the bail, which was termed temperament, was given a score from the following scale 1) docile, 2) slightly restless, 3) restless, 4) nervous, 5) wild, 6) aggressive. Grandin (1993), in a comparable subjective temperament test, restrained individuals in a squeeze chute and rated their reaction based on a similar scale: 1) calm, no movement 2) slightly restless 3) squirming, occasionally shaking squeeze chute 4) continuous, very vigorous movements and shaking of the squeeze chute 5) rearing, twisting of the body and struggling violently. Variations on this subjective scoring method have been used extensively.

In an intensive investigation of the effects of frequent handling on the temperament of cattle, Boissy & Bouissou (1988) performed a variety of tests on dairy heifers that is typical of many of the cattle temperament studies. One group of tests involved interactions with humans, while the other did not. The non-human tests included an open field test, a subjective rating of the heifers' reactions towards a novel barrier across an alleyway, and a timed feeding test in a novel environment; these tests were designed to evaluate the heifers' general anxiety and reactivity towards novelty. The tests involving humans consisted of a timed feeding test near a human, a flight distance test in which the distance to which a handler could approach before the heifer moved away was recorded, a time to capture test, and a subjective evaluation of the heifers' reactions to being lead on a halter. All of these behaviour measures combined to give a general impression of the heifers' reaction towards handling. Several other studies have used a similar battery of tests, with slight variations (Le Neindre *et al.* 1995; Gringard *et al.* 2001; Van Reenen *et al.* 2004; Kilgour *et al.* 2006).

Another measure used to evaluate temperament in cattle is the flight speed test developed by Burrow *et al.* (1998), where an animal's time to cover a set distance is recorded after release from some form of restraint, generally a chute or scale. Several other researchers have used the flight speed test, with slight variations in the distance traveled, *etc.* (Fell *et al.* 1999; Tõzér *et al.* 2003; Curley *et al.* 2006; Müller *et al.* 2006; Kilgour *et al.* 2006). In this test, a high flight speed *i.e.* a shorter time to cross the set distance, is considered indicative of a more excitable or reactive animal.

Stookey *et al.* (1994) developed a device that objectively quantifies an animal's reaction to handling. The movement measurement device (MMD) operates in conjunction with a digital scale, and records a peak when a trend of an increasing or decreasing load on the scale is reversed. A higher number of peaks is indicative of more movement by the animal, and thus a higher reactivity to the test situation. The MMD has been subsequently used in a quantitative trait loci (QTL) mapping study of temperament in cattle (Schmutz *et al.* 2001) and to quantify cattle's behavioural response to noise (Waynert *et al.* 1999).

Another device to objectively measure cattle behaviour was reported by Schwartzkopf-Genswein *et al.* (1997). They used a series of several load cells and strain gauges at various points on a squeeze chute and head gate to evaluate the forces exerted by steers as a reaction to restraint and branding procedures.

Temperament studies conducted to date in smaller ruminants such as sheep and goats have generally relied on measurements very similar to those used in cattle. For example, Romeyer & Bouissou (1992) and Vandenheede & Bouissou (1993) used open field tests, and subjective observation of reactions to the sudden appearance of novel objects or humans, to judge the general reactivity and temperament of sheep. However, in contrast to rodent studies, Romeyer & Bouissou (1992) considered increased general activity in the open field as a measure of increased fear. Lyons (1989), in a study of dairy goats, used subjective ratings of reactions to a human in their home pen, as well as ratings of 7 traits (excitable, tense, watchful, apprehensive, confident, friendly to people, and fearful of people) during milking to determine overall temperament.

In pigs, most researchers tend to investigate "personality" rather than "temperament" (Forkman *et al.* 1995). Two of the common methods for evaluating personality in pigs are the social confrontation test and the back test. Social confrontation tests involve placing an unfamiliar pig into another pig's home pen, observing the resulting fights, and classifying the resident pig as either aggressive or non-aggressive (Hessing *et al.* 1993). The back test involves restraining a piglet on its back and recording the number of escapes attempted, > 2 attempts being classified as a resistant piglet, 2 attempts as intermediate, and < 2 being a non-resistant piglet (Hessing *et al.* 1993). Vocalizations in pigs are also recorded, as they are considered to be indicative of stress (Schrader & Todt 1998; Désautés *et al.* 1997). Pigs are also subjected to open field and response to novel object type tests (Désautés *et al.* 1997).

Open field behaviour has been used extensively to measure fear related behaviours in chickens (Buitenhuis *et al.* 2004) and Japanese quail (*Coturnix japonica*) (Miller *et al.* 2005). In addition to movement measurements, the number and type of vocalizations are also often measured in chickens (Buitenhuis *et al.* 2004). A test that is specific to poultry is the tonic immobility test, where fearful birds will remain motionless after restraint on their backs. This test is used in chickens (Jensen *et al.* 2005) and quail (Minvielle *et al.* 2002), and the duration of the tonic immobility and the number of restraint periods needed to induce tonic immobility are generally recorded. The inability to induce tonic immobility (or a large number of required attempts), and a short duration of tonic immobility are considered to represent reduced fearfulness.

2.1.3 Associations with production

Numerous studies have been conducted in cattle and other livestock species that demonstrate associations between favorable temperament and various measures of productivity.

2.1.3.1 Other livestock

In a study of 16 dairy goats, Lyons (1989) examined temperament for associations with the inhibition of milk let down. The goats were given subjective temperament ratings by two separate observers after 21 to 35 days of twice daily milking. Objective measurements of the goats' reaction to a human in their home pen were also taken. Inhibition of milk let down was measured by first milking the goats by machine, then collecting any remaining milk by hand, and finally giving an oxytocin injection to release any retained milk; the amount of residual milk was then expressed as a percentage of the total milk collected. A consistent relationship was found, with the calmer, less reactive goats having a significantly lower inhibition of milk let down as compared to the nervous, reactive goats.

Hessing *et al.* (1994) examined the production effects of housing grow/finish pigs in groups based on their individual personality traits. Two back tests were conducted, and based on the results the piglets were classified as either resistant (R; n=86), non-resistant (NR; n=94) or doubtful (D; n=26). Piglets were then placed into pens of all R pigs (R/R), all NR pigs (NR/NR) or mixed groups of R and NR pigs (R/NR). Aggressive behaviour after grouping was highest in the R/R pens. At the end of the finishing period, carcass weight, meat %, and ADG were all significantly better for pigs in the R/NR pens than in the R/R or NR/NR pens. Specifically, ADG in the R/NR pens was 40g/day higher than in R/R pens and 28g/day higher than in NR/NR pens.

In an investigation into whether selection for behavioral traits affects production in Japanese quail, Minvielle *et al.* (2002) studied two lines of quail which had previously been selected for long and short tonic immobility responses as well as a line which had been selected solely for early egg production. The tonic immobility test results showed significant differences between the three quail lines, with high egg production line having values intermediate to the long and short tonic immobility lines. Egg production was not significantly different between short tonic immobility and high egg producing lines, but was significantly lower in the long tonic immobility line. Growth traits for the high egg producing line were intermediate between the long and short tonic immobility lines, with short tonic immobility showing the highest growth rates.

In a simple test of broiler chick fearfulness towards humans, Hemsworth *et al.* (1994) recorded the number of chicks that remained close to or oriented towards a human walking in their midst. This test was performed on 22 farms, and the results showed that feed conversion was significantly lower on farms where the chicks actively avoided humans.

Marín *et al.* (1997) conducted another test of broiler chick performance in relation to fear behaviour. They used a T-maze apparatus to test two day old chicks' anxiety in a novel environment and their response to isolation. Chicks with a short time to navigate the maze were considered to be less anxious, and designated high performance (HP), while chicks that did not escape the maze were considered low performance (LP). At fifteen days of age, male HP chicks had 12.7% higher body weight than male LP chicks, and a similar, but non-significant, trend was observed in the female HP and LP chicks.

Schütz *et al.* (2004) examined the relationship between performance and fear behaviour in laying hens by observing the F2 generation of a Red Jungle fowl X White Leghorn intercross. Production parameters measured included growth, age at sexual maturity, feed intake and egg production. Behaviour measures included tonic immobility, open field, novel object and response to restraint tests. Hens with the highest growth rates showed the lowest total movement in the open field test and shortest times to explore novel food, while in males high growth rates were associated with longer tonic immobility. While these results seem to indicate that behaviours typically associated with fearfulness are also associated with higher growth, the authors argue that higher growth could simply be a reflection of a higher feeding motivation in these birds as indicated by the short time to explore novel foods. More expected results were that higher egg weight was associated with higher open field activity, and the trend that the behavioural responses to stress in high producing F2s were more similar to that of the domestic Leghorns than to the wild Red Jungle fowl parents.

2.1.3.2 Cattle

Several researchers have shown a relationship between temperament in cattle and growth traits. Tulloh (1961) reported that animals with lower subjective temperament scores (more favorable) had significantly higher live weights than those with higher scores.

Voisinet *et al.* (1997a) studied temperament in 436 beef cattle of several breeds and crosses in a feedlot environment. The cattle were weighed at 28 day intervals, and two independent, subjective temperament observations were made. The first observation was collected during the animals' first experience in the handling facility while the cattle were restrained in a squeeze chute and head gate; temperament scores were given on a scale of 1 to 4. The second observation was collected after the cattle had 4 to 8 experiences in the handling facility, and while they were in a non-restraining scale; temperament scores were given, by a different observer, on a scale of 1 to 5. For both observations, cattle with the calmest temperaments (lowest score) had the highest ADG.

Fell *et al.* (1999) also examined the relationship between feedlot performance and temperament in cattle. After a flight speed test, a subjective temperament score and a novel object test, 12 animals with extremely poor temperament and 12 with extremely good temperament were selected as focal animals for a study of feedlot performance. The cattle from the poor temperament group had significantly lower ADG after 85 days on feed than the cattle from the good temperament group.

In a similar study, Petherick *et al.* (1997) grouped *Bos indicus* x *Bos taurus* steers based on flight speed results into “poor” and “good” temperament groups; they showed a trend for the poor temperament group to have lower ADG than the good temperament group. In a later study, Petherick *et al.* (2002) grouped *Bos indicus* steers into poor, mixed and good temperament groups based on flight speed tests, and found that there was a trend for the steers in the poor group to have lower ADG than the good group. They also report that there was a significant negative correlation between flight speed values and both ADG and body condition score.

In an interesting contrast to the above studies, Müller *et al.* (2006) showed that in cross bred Angus heifers, flight speed showed a quadratic relationship with ADG, as opposed to the simple linear relationship which other authors had previously described.

Many studies have also been conducted evaluating the relationship between cattle temperament and meat quality, although the results are somewhat inconsistent. For example, Voisinet *et al.* (1997b) showed that *Bos indicus* cross steers with the higher subjective temperament scores had a higher incidence of borderline dark cutting carcasses, as well as higher Warner-Bratzler Shear force (WBSF) values indicative of tougher meat. Kadel *et al.* (2006) studying *Bos taurus,* and King *et al.* (2006) studying *Bos indicus* showed associations of lower flight speeds with lower WBSF. King *et al.* (2006) also failed to find any associations between temperament and dark cutting. Also interesting is that Petherick *et al.* (1997) report that there were no differences in any carcass traits between *Bos indicus* x *Bos taurus* steers grouped by flight speed.

Petherick *et al.* (2002) report that flight speed was significantly negatively correlated with dressing percentage, and that there were indications of poor temperament being associated with lower initial muscle pH and increased heat shortening of the muscle.

2.1.4 Genetics of fear/anxiety response

There is significant evidence that temperament has genetic components. Behaviour varies both between (Tulloh 1961; Stricklin *et al.* 1980) and within (Le Neindre *et al.* 1995) specific breeds of cattle and chickens (Jensen *et al.* 2005), and many lines of poultry and rodents have been selected based on divergent behaviours. Behavioural variations have also been associated with specific phenotypes such as colour in cattle (Watts & Stookey 2001; Tõzér *et al.* 2003). The following section will briefly review some of the work conducted to date regarding the genetics of temperament and other fear/anxiety responses.

2.1.4.1 Rodents

The genetics of fear/anxiety related behaviours has been studied extensively in rodents. In fact, in Flint's (2003) review of animal behaviour QTL studies, 86 separate loci for "emotionality" components in rodents were listed. A few of these studies will be examined here.

Ramos *et al.* (1998), using crosses of Lewis (LEW) and spontaneously hypertensive (SHR) rats, demonstrated that difference in elevated plus maze and open field behaviour between the two strains was due to direct genetic effects, and that central locomotion was the most heritable component of these behaviours.

In a QTL study of elevated plus maze and open field behaviour in LEW x SHR rats, Ramos *et al.* (1999) identified a significant QTL for central open field locomotion on chromosome 4 in female rats. They suggested Tac1r, which encodes a neurotransmitter receptor, as a candidate gene for this QTL.

Gershenfeld *et al.* (1997) used crosses between A/J and C57BL/6 mice, which differ significantly in open field behaviour, in a QTL study of a variety of open field responses. Significant QTL for initial movement and vertical movements in the open field were located on chromosomes 1 and 10, respectively. No candidate genes were reported for either QTL.

2.1.4.2 Cattle & Other livestock

Temperament has been shown to be variable between breeds of cattle. Tulloh (1961) studied Hereford, Angus and Shorthorn cattle, and concluded that Herefords were the most docile, while the Shorthorns were the least docile. Stricklin *et al.* (1980) used a subjective rating of temperament to determine that British cattle breeds were more docile that other European breeds, with Herefords showing the most docile temperaments overall. Gauly *et al.* (2002) found a similar result, with Simmentals showing more agitation in response to handling than Angus.

Similar variations in fear/anxiety related behaviours have also been observed between breeds in other species of livestock. Romeyer & Bouissou (1992) used open field, novel object, surprise and presence of a human tests to concluded that Ile-de-France sheep were generally more fearful than Romanov sheep. Désautés *et al.* (1997), in a study of Meishan x Large White pigs, concluded that differences in piglets' locomotion and vocalization in a novel environment were due to the breed differences.

Various studies have examined the heritability of temperament in beef cattle. Stricklin *et al.* (1980) reported the heritability of temperament as 0.44±0.18 in 388 crossbred calves, and 0.48±0.29 in 243 purebred bulls. Le Neindre *et al.* (1995), in a study of Limousin cattle, calculated the heritability of docility in heifers as 0.22. Schmutz *et al.* (2001), using the MMD to objectively rate response to handling, calculated the heritability of temperament at weaning as 0.36 in a study of cattle embryo transfer families. Kadel *et al.* (2006) calculated the heritability of flight time as 0.30 at weaning, and as 0.36 at finishing in *Bos indicus* breeds.

Several QTL studies have been performed in livestock in regards to fear/anxiety behaviours. In a study of embryo transfer families in beef cattle, Schmutz *et al.* (2001) identified 6 QTL related to temperament, located on chromosomes 1, 5, 9, 11, 14 and 15. In a study of temperament in dairy cattle, Hiendleder *et al.* (2003) found a QTL on chromosome 29 for reaction to milking.

Beaumont *et al.* (2005) studied crosses between lines of quail that had been selected for long and short tonic immobility. Significant QTL were found on linkage group 1 for log tonic immobility and the number of induction attempt required to induce tonic immobility, and on linkage group 3 for the number of jumps and defecations in the open field test.

Schütz *et al.* (2004) used Red Jungle fowl x White Leghorn crosses to identify QTL associated with stress behaviours and production. Significant QTL for tonic immobility and novel object responses were found in males on chromosome 1, in regions of previously identified QTL for growth. However, three other significant QTL for stress behaviours were not associated with production traits.

In a QTL study of open field behaviour in chickens, Buitenhuis *et al.* (2004) examined crosses between two White Leghorn lines that had been selected for divergent levels of feather pecking. The birds were tested at 5 and 29 weeks of age, and a different QTL associated with fear parameters was reported for the two tests; although both fear QTL were on chromosome 4, they were not located in the same region.

Two studies have also looked for QTL for feather pecking in chickens, as this behaviour has been shown to be associated with other stress response behaviours. Buitenhuis *et al.* (2003), in a study of crosses between lines of White Leghorns selected for divergent levels of feather pecking, reported a significant QTL for severe feather pecking on chromosome 2, a suggestive QTL on chromosome 10 for gentle pecking in 6 week old birds, and two suggestive QTL on chromosomes 1 and 2 for gentle pecking in 30 week old birds. Jensen *et al.* (2005) identified a suggestive QTL for feather pecking on chromosome 3 in Red Jungle fowl x White Leghorn crosses.

With the exception of Schmutz *et al.* (2001), none of these QTL studies have proposed candidate genes for any of the relationships discussed. Schmutz *et al.* (2001) suggested cannabinoid receptor 1 (CNR1) as a candidate gene for the QTL on cattle chromosome 9, and type 2 dopamine receptor (DRD2) as a candidate gene for the QTL on cattle chromosome 15.

2.1.5 Physiology of fear/anxiety response

Neuroendocrine responses to stress, through both the sympathetic adrenal medullary system and the hypothalamic-pituitary adrenal (HPA) axis, and their link with behaviour, have been well documented (Castanon & Mormède 1994; Boissy 1995). The following section will briefly review the HPA axis, followed by a review of several studies that have used measures of HPA axis activity or other physiological parameters to study fear/anxiety response behaviour.

2.1.5.1 The HPA Axis

Corticotropin releasing hormone (CRH) is one of the key hormones in the HPA axis response to stress (Miller & O'Callaghan 2002). The processes involved in the HPA axis stress response are as follows: upon the detection of a stressor by various sensory systems, the hypothalamus is stimulated and releases CRH, which in turn stimulates CRH receptors in the pituitary triggering increased adrenocorticotropin (ACTH) production and release; once in the circulation ACTH acts on the adrenal gland through melanocortin receptor-2 and causes the release of cortisol in most mammals (or corticosterone in rodents) which serves to buffer tissue damage and also inhibits further CRH and ACTH stimulation of the HPA axis (Miller & O'Callaghan 2002; Slominski *et al.* 2000). The HPA axis stress response pathway is summarized in Figure 2.1.

2.1.5.1.1 Physiology studies

Many studies of behavioural response to fear producing stressors in livestock measure physiological parameters, with plasma cortisol levels (representing HPA axis activity) being the most commonly reported measure.

Boissy & Bouissou (1988) examined differences in heifers' behavioural and physiological responses to a variety of stressors between groups exposed to different amounts of handling by humans. The group of heifers which had received the handling treatment over the longest period showed significantly lower heart rate and plasma cortisol in response to handling, and were generally less agitated around humans than the groups which had received the same number of handling treatments over a shorter interval.

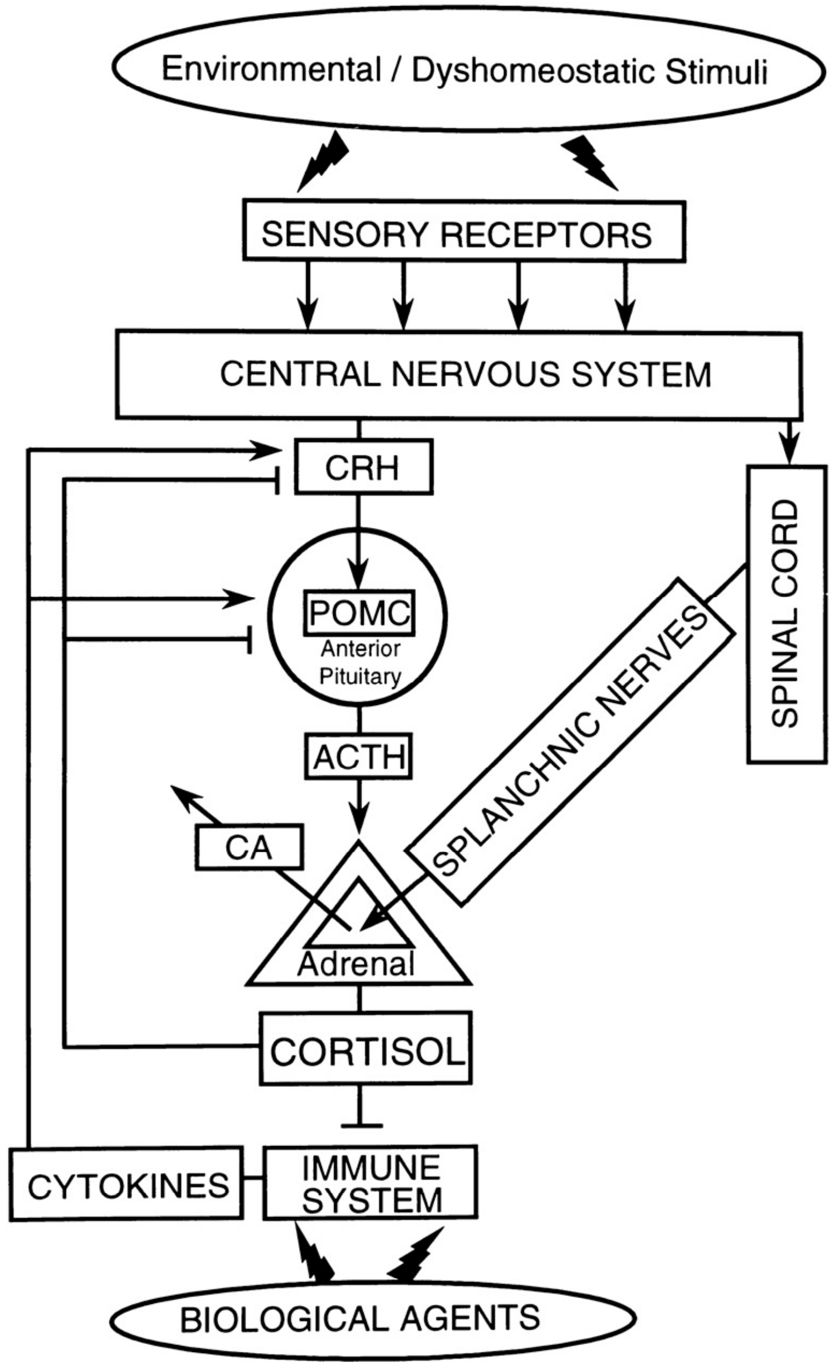


Figure 2.1 The hypothalamic-pituitary adrenal axis stress response pathway (Slominski *et al.* 2000). CRH=corticotropin releasing hormone, POMC=pro-opiomelanocortin, ACTH=adrenocorticotropin, CA=catecholamine

When Gauly *et al.* (2002) examined the relationship between plasma cortisol levels and behavioural indicators of stress in calves exposed to a restraint test, they found that both cortisol levels and agitation were higher in Simmental as compared to Angus calves, and in female as compared to male calves.

Van Reenen *et al.* (2005) found consistent relationships between high plasma cortisol levels and avoidance of novel objects. However, they did not find any consistent relationships between cortisol levels and open field behavior results, or between heart rate and any of the behaviour tests. Curley *et al.* (2006) showed a positive correlation between exit velocity and subjective pen scores with serum cortisol levels in yearling Brahman bulls.

Bristow & Holmes (2007) not only examined the relationship between cortisol and behaviour, but also examined whether cortisol levels were predictive of cattle behaviour. They studied a total of 9 Angus x Hereford cows under grazing and restrained conditions, and found the cows with higher baseline cortisol levels spent less time ruminating and tended to have increased vocalizations and remain closer to other cows while grazing.

Similar studies, examining behavioural and endocrine stress responses, have also been performed in swine. Désautés *et al.* (1997) examined Meishan and Large White pigs and their crosses. They found that Large White pigs were more active than Meishans in open field tests, and that Large Whites had lower post-stress cortisol than Meishans. These results are somewhat odd, as Meishans are generally considered to be very calm compared to Large Whites, and therefore one would expect that Meishans would have more activity in the open field and lower cortisol than Large Whites. However, the authors argue that Meishans may simply have an extremely low motivation for exploration and a very passive system of coping with stress (thought to be associated with high cortisol by some researchers).

Schrader & Todt (1998) studied the relationship between vocalizations in pigs during isolation and restraint, and found that a decrease in the number of grunt vocalizations was significantly associated with an increase in plasma cortisol levels.

In a study of stress responses in chickens, Marín & Jones (1999) showed that chicks that had been scored as high performing in a T-maze test (indicative of low fearfulness) also had significantly lower plasma cortisol levels compared to low performing chicks (high fearfulness) in response to being partially submerged in water.

Manteca & Deag (1993) discussed problems associated with using physiological measures such as plasma cortisol to interpret stress reactivity in animals. Several factors need to be considered when interpreting cortisol results, as levels can vary with daily rhythms or be affected significantly by an animal's previous experiences and interpretation of the test situations. They stated that learning in a novel environment may increase plasma cortisol, and therefore increased cortisol levels may not be indicative of adverse reactions to stress in that situation. They also explored the active versus passive coping strategy theory, which states that animals with an active coping style will show aggression, increased activity and an increased heart rate in response to stress, while animals with a passive coping style will show an increase in plasma cortisol accompanied by a decrease in heart rate and locomotion. The authors conclude that cortisol levels alone cannot be used to interpret fearfulness, etc., but rather that they should always be used in conjunction with other behaviour measurements.

2.2 CRH

The existence of a substance in the hypothalamus that increases corticotropin secretion was demonstrated in 1955 by both Guillemin & Rosenberg (1955), and by Saffran & Schally (1955). The mature 41 amino acid corticotropin-releasing hormone (CRH) peptide was first isolated, sequenced and synthesized by Vale *et al.* (1981) from ovine hypothalamus tissue.

2.2.1 *CRH* Gene structure and location

Furutani *et al.* (1983) and Shibahara *et al.* (1983) used the amino acid sequence published by Vale *et al.* (1981) to sequence and characterize ovine and human prepro-CRH, respectively. Furutani *et al.* (1983) showed that the ovine prepro-CRH protein consists of 190 amino acids, including a 24 amino acid signal peptide.

Shibahara *et al.* (1983) determined that the human CRH gene (*CRH*) consists of two exons separated by an 800 bp intron, with the entire CRH coding region in the second exon. The human prepro-CRH protein consists of 196 amino acids (Figure 2.2), also with a 24 amino acid signal sequence, and with 7 amino acid substitutions in the mature peptide as compared to the ovine sequence (Shibahara *et al.* 1983). Rat (Thompson *et al.* 1987) and ovine (Roche *et al.* 1988) *CRH* also consist of two exons with the coding region in exon 2, as do all other *CRH* reported thus far.

When Mimmack *et al.* (1998) amplified porcine *CRH*, they determined that the porcine coding sequence was 80% identical to the human sequence, and 84% and 85% identical to sheep and rat sequences respectively; the porcine amino acid sequence is 83%, 74% and 75% identical to the human, sheep and rat amino acid sequence, respectively. The region coding for the hormone product is highly homologous across species (Mol *et al.* 1994). A highly conserved ~350 bp promoter region containing numerous response elements has also been identified immediately upstream of the 5' UTR (King *et al.* 2001; King & Nicholson 2007).

Arbiser *et al.* (1988) were the first to map *CRH* when they used *in situ* hybridization to map human *CRH* to chromosome 8q13. Further work has mapped *CRH* to the following locations: mouse chromosome 3 (Knapp *et al.* 1993), sheep chromosome 9 (Broad *et al.* 1995), cattle chromosome 14 (Barendse *et al.* 1997), rat chromosome 2 (Laes *et al.* 2001), goat chromosome 14 (Pinton *et al.* 2000), and pig chromosome 4q13 (Pinton *et al.* 2000; Wimmers *et al.* 2002).

```
Homo sapiens          MRLPLLVSAG VLLVALLPCP PCRALLSRGP VPGARQAPQH PQPLDFFQPP 50
Pan troglodytes       MRLPLLVSAG VLLVALLPCP PCRALLSRGP VPGARQAPQH PQPLDFFQPP 50
Canis familiaris      MRLPLLLSAG VLLVVSLPCP PCRALLSRGP IPGARQAAQH PQPLDFFQLP 50
Bos taurus            MRLPLLVSVH VLLVALLPSP PCRALLSRGP IPGARQASQH PQPLXFFQPP 50
Mus musculus          MRLRLLVSAG MLLVALSSCL PCRALLSRGS VPRAPRA--- PQPLNFLQ-P 46
Gallus gallus         MKLQPLVCAG ILLLALLPCH ECRALSKS-- -PFAARG--A LQQPDFFPQQ 45
                      *:*  *:..* :**:.  ..   **** .     . * :.     *   *:

Homo sapiens          PQSEQPQQPQ ARPVLLRMGE EYFLRLGNLN KSPAAPLSPA SSLLAGGSGS 100
Pan troglodytes       PQSEQPQQPQ ARPVLLRMGE EYFLRLGNLN KSPAAPLSPA SSLLAGGSGS 100
Canis familiaris      PQPQQPQQPQ ARPVLYRMGE EYFLRLGNLN KSPAAPLLPA SSPVAGGSGS 100
Bos taurus            PQPQEPQ--- ALPTLLRVGE EYFLRLGNLD ETRAAPLSPA ASPLASRSSS 97
Mus musculus          EQPQQPQ--- --PVLIRMGE EYFLRLGNLN RSPAARLSPN STPLTAGRGS 91
Gallus gallus         QQQQQQQ--- TLPVLLRMGE EYFLRLGHLT KRPAG----- --PFSASSGG 85
                       * :: *  * * *:** *******:*  .   *.        .:.  ..

Homo sapiens          RPSPEQATAN FFRVLLQQLL LPRRSLDSPA ALAERGARNA LGGHQEAPE- 149
Pan troglodytes       RPSPEQATAN FFRVLLQQLL LPRRSLDSPA ALAERGARNA LGGHQEAPE- 149
Canis familiaris      RLSPDEAAAN FFRALLQQLP LPRRQLDSPA GPAERGEENA LGSRQETPE- 149
Bos taurus            RLSPDRVAAN FFRALLQ--- -PRRPFDSPA GPAERGTENA LGSTQEAPAA 143
Mus musculus          RPSHDQAAAN FFRVLLQQLQ MPQRSLDSRA EPAERGAEDA LGGHQGALE- 140
Gallus gallus         HLRP-EASAE LLRAAAAQLQ ------GSGS PEGDEGAG-- -----EAVE- 120
                      :    :.:*: ::*.             .* :   .:.*           :

Homo sapiens          RERRSEEPPI SLDLTFHLLR EVLEMARAEQ LAQQAHSNRK LMEIIGK 196
Pan troglodytes       RERRSEEPPI SLDLTFHLLR EVLEMARAEQ LAQQAHSNRK LMEIIGK 196
Canis familiaris      RERRSEEPPI SLDLTFHLLR EVLEMARAEQ LAQQAHSNRK LMEIIGK 196
Bos taurus            RERRSQEPPI SLDLTFHLLR EVLEMTKADQ LAQQAHINRK LLDIAGK 196
Mus musculus          RERRSEEPPI SLDLTFHLLR EVLEMARAEQ LAQQAHSNRK LMEIIGK 187
Gallus gallus         REKRSEEPPI SLDLTFHLLR EVLEMARAEQ LAQQAHSNRK LMEIIGK 167
                      *::**:**** ********** *****::*:* ****** *** *::* **
```

Figure 2.2 Multiple sequence alignment for CRH, generated from using ClustalW software (Chenna *et al*. 2003). Identical positions are indicated with "*", conserved substitutions with ":", semi-conserved with ".", and gaps with "-". The 41 a.a. CRH hormone is indicated in bold text. NCBI reference numbers for the sequences used are (from top to bottom): NP_000747.1, XP_519792.2, NP_991338.1, NP_001013418.1, NP_991338.1 and XP_418279.1

2.2.2 *CRH* Gene expression

Corticotropin-releasing hormone has been found in a variety of tissues by radioimmunoassay (RIA). Suda *et al.* (1984) used RIA in humans to find *CRH* in the following brain regions: hypothalamus, pituitary, thalamus, cerebral cortex, cerebellum, pons, medulla oblongata and spinal cord. They also demonstrated the presence of *CRH* in lung, liver, pancreas, duodenum and adrenals. Kawai *et al.* (1985) performed a similar analysis in sheep, and found *CRH* in the above mentioned tissues as well as in stomach, jejunum, and ileum. Muglia *et al.* (1994) added to the knowledge of *CRH* expression by confirming its presence in mouse heart, ovaries and testes.

Shibasaki *et al*. (1982) showed that a compound extracted from human placental tissue had CRH-like effects on rat pituitary cell cultures. Subsequently, Robinson *et al.* (1989) found *CRH* in human, gorilla and rhesus monkey placenta, but not in placenta from lemurs, rats or guinea pigs.

The main factor regulating *CRH* expression is generally considered to be adrenal glucocorticoids (cortisol or corticosterone) acting in a classical negative feedback loop. However, it has been shown that the effect of glucocorticoids on *CRH* expression differs in various brain tissues: corticosterone decreases *CRH* mRNA in the paraventricular nucleus of the hypothalamus, but increases *CRH* mRNA in the amygdala (Watts & Watts-Sanchez 1995). Watts (2005) concluded that the regulation of *CRH* expression by glucocorticoids is far more complex than is indicated by the classic negative feedback model. It has also been shown that other stress related hormones such as noradrenaline affect *CRH* expression (Itoi *et al.* 1998; Itoi *et al.* 2004).

2.2.3 Effects of CRH

As was previously briefly discussed, CRH is one of the main activators of the HPA axis stress response pathway. However, the physiological effects of CRH are not limited to its direct role in the HPA axis, although they generally consist of stress adaptation mechanisms. Dunn & Berridge (1990) conducted an extensive review of the effects of CRH administration, which include endocrine alterations such as inhibition of luteinizing hormone and growth hormone secretion, increased plasma adrenaline and noradrenaline, activation of the sympathetic nervous system, and various gastrointestinal alterations.

The various well documented behavioural effects of CRH warrant the most discussion for the purposes of this review. CRH has reported effects on locomotion under stress, sexual behaviour, grooming, feeding and learning, with a general pattern that CRH increases anxious responses (reviewed by Dunn & Berridge 1990; Heinrichs *et al.* 1995; Deussing & Wurst 2005).

Particularly interesting are the studies that have utilized mice that over-express *CRH* or *CRH* knock-outs. Stenzel-Poore *et al.* (1994) found that mice over-expressing *CRH* show increased anxiety in elevated plus maze and open field tests as compared to their normal siblings. Groenink *et al.* (2003) after a review of similar studies done by other groups, concluded that the results were too inconsistent to simply conclude that over-expressing *CRH* definitively leads to increased anxiety. In a *CRH* knock-out study, Muglia *et al.* (1995) observed that CRH-/- mice were identical in appearance and general behaviour to their normal siblings, although they had significant histological and immunohistochemical differences.

2.2.4 Mutations in *CRH*

Because of the myriad effects of CRH, several researchers have attempted to find correlations between various mutations in *CRH* and human pathologies or livestock production traits. Polymorphisms in the *CRH* promoter have been implicated in rheumatoid arthritis in humans (Baerwald *et al*. 1997), and the *CRH* system has been examined extensively in clinical depression and other psychiatric disorders (Villafeurte *et al. 2002;* Liu *et al.* 2006).

In livestock, the *CRH* system has been examined primarily due to the effects of stress on energy partitioning and therefore growth, as well as the more direct anorexic effects of CRH. In addition to the cattle studies discussed below, some work has also been performed with swine. Muráni *et al.* (2006a) discussed *CRH* as a positional and functional candidate gene for QTL for several carcass and growth traits in a German swine research herd. Muráni *et al.* (2006b) subsequently showed that a SNP (single nucletotide polymorphism) in the *CRH* promoter region was associated with a few growth and carcass traits in German commercial swine strains, although they concluded that this particular SNP did not have a significant overall effect on growth.

2.2.4.1 Mutations in cattle

Buchanan *et al.* (2000) identified two growth QTL on cattle chromosome 14, and proposed *CRH* as a positional candidate gene for one of the QTL. Subsequent work identified a SNP at nucleotide 240 in exon 2 of *CRH*; a C>G substitution resulting in a non-conserved amino acid change at residue number 77 from a histidine to aspartic acid (Buchanan *et al.* 2002b). The C allele of this *240C>G* SNP showed positive correlation with the weaning weight and yearling weight EPDs of 429 beef bulls.

Further work by the same research group identified 2 additional novel SNPs in *CRH*. They examined all 3 *CRH* SNPs and previously reported SNPs in *LEP*, *MC4R* and *POMC* for associations with growth and carcass traits in 256 Charolais cross steers (Buchanan *et al.* 2005). The C>G substitution at residue 4 (*22C>G*) causes the fourth amino acid in the *CRH* signal sequence to change from a proline to an arginine. There was a significant effect of the *CRH 22C>G* SNP for end-of-test rib-eye area and hot carcass weight, with the GG animals having increased values compared to *CC* animals. The *CRH 240C>G* SNP had a significant effect for hot carcass weight. In addition, there were significant interactions between the *CRH 22C>G* SNP and the *POMC* and *MC4R* SNPs for end-of-test rib-eye area and hot carcass weight, respectively.

2.3 LEP

Leptin is the hormone product of the obese gene, which was originally characterized in mice by Zhang *et al.* (1994). This discovery spurred a prodigious amount of research, largely in hopes of discovering a genetic basis for human obesity. For the purposes of the remainder of this review, the gene coding for leptin will be referred to as *LEP*.

2.3.1 *LEP* Gene structure and location

Zhang *et al.* (1994) localized mouse *LEP* to chromosome 6, and Green *et al.* (1995) mapped it to human 7q31. Pomp *et al.* (1997) confirmed the work of Stone *et al.* (1996) in determining that *LEP* is located on cattle chromosome 4.

Zhang *et al.* (1994) also determined that mouse *LEP* consists of 3 exons, with the coding regions occurring on exons 2 and 3, that there is a 21 amino acid signal sequence and that the mature peptide is 167 amino acids in length (Houseknecht & Portocarrero 1998). Houseknecht & Portocarrero (1998) reported that *LEP* is highly conserved in vertebrates, and that sequence homology between humans, rats, mice, cattle, pigs and chickens ranges from 83% to 97%.

2.3.2 *LEP* Gene expression

Originally, *LEP* expression was reported only in white adipose tissue (Zhang *et al.* 1994). However, further research has shown *LEP* expression in a variety of other tissue such as brown adipose tissue (Moinat *et al.* 1995), placenta, and fetal bone (Hoggard *et al.* 1997), mammary gland epithelial cells (Smith-Kirwin *et al.* 1998) and in stomach (Bado *et al.* 1998). Expression of *LEP* is increased by insulin, glucocorticoids, estrogen and glucosamine, and is decreased by adrenaline and noradrenaline (Rayner & Trayhurn 2001).

2.3.3 Physiological effects of LEP

The most well known effect of *LEP* is on the regulation of appetite and body weight. Leptin, secreted from adipocytes, acts on various neuropeptides including neuropeptide Y(NPY), melanin concentrating hormone, POMC, melanocortin stimulating hormone (α-MSH), agouti-related peptide, galanin, CRH, and ACTH among others to influence food intake and thus regulate body weight (Ingvarsen and Boisclair 2001).

Leptin has significant effects on reproduction (Cervero *et al.* 2006) and immune response (Matarese *et al.* 2005). Leptin also has an effect on bone formation (Pogoda et al. 2006). However, for the purposes of this review, the effects of leptin on the HPA axis and cortisol are the primary interest.

Bornstein *et al.* (1997) showed that leptin has a direct effect on cortisol secretion in adrenocortical cell cultures, with increasing leptin dosage resulting in a decrease in cortisol secretion. In a study of the effects of leptin administration on HPA axis activation due to restraint stress in mice, Heiman *et al*. (1997) found that leptin treated mice had lower plasma corticosterone and ACTH levels after restraint than did the controls. Heiman *et al*. (1997) concluded that the definition of the HPA axis should be expanded to include the effects of leptin.

The link between disturbances in the HPA axis and psychiatric disorders in humans such as depression is well documented (Kasckow *et al.* 2001) and therefore factors that affect the

HPA axis, including leptin, have been investigated as potential treatments for such disorders. Lu *et al.* (2006) showed that rats subjected to chronic unpredictable stress had lower leptin and higher corticosterone levels than the controls. Lu *et al.* (2006) also showed that leptin administration resulted in behavioural responses in forced swimming that are very similar to those seen with the administration of a common anti-depressant.

2.3.4 Mutations in *LEP*

Because leptin has such a clear influence on obesity and other important physiological traits, many researchers have conducted association studies with various polymorphisms in leptin. For example, Stobel *et al*. (1998) showed an association between a *LEP* SNP and obesity in humans, and Jiang & Gibson (1999) examined four *LEP* SNPs for associations with fat traits in swine.

2.3.4.1 Mutation in cattle

Fitzsimmons (1999) identified a single nucleotide polymorphism (SNP) in the fourth amino acid of the mature leptin peptide, which is a C>T transition that changes the amino acid produced from arginine to cysteine (*73C>T*). Buchanan *et al*. (2002a) found that homozygous T animals produced higher levels of *LEP* mRNA and that the T allele was associated with higher levels carcass fat. Buchanan *et al.* (2003) found that Holstein cows homozygous for the T allele had higher milk and milk protein yield. Buchanan *et al.* (2007) conducted further work on the effects of this SNP, and again showed strong association between the T allele and various fat measurements. Additionally, Buchanan *et al.* (2007) found that Charolais cross steers that were TT for the SNP had higher serum leptin levels than CC Charolais steers when they were finished to a backfat depth of 12 mm.

In a study with the same *LEP* SNP, Kononoff *et al*. (2005) showed that the TT genotype was associated with a higher percentage of carcasses grading Canada AAA (related to marbling), and a lower proportion of carcasses of Canada Yield Grade 1 (related to % lean meat yield).

2.4 Objective & Hypothesis

The purpose of this thesis was to determine if there were any associations between SNP in two different genes and temperament in beef cattle. The three SNPs examined, one in Leptin and two in CRH, had been previously studied in relation to beef carcass and growth traits (Buchanan *et al.* 2002a; Buchanan *et al.* 2002b; Buchanan *et al.* 2005). Leptin and CRH are both involved in the HPA stress response axis; add to this the previous associations with growth and carcass traits, and Leptin and CRH become interesting candidates for associations with temperament. Our hypothesis was that steers with the *LEP TT* genotype would have the calmest temperaments.

3 MATERIALS & METHODS

3.1 Animals & Processing

This experiment was conducted using 400 crossbred beef steers purchased from an auction market located near Saskatoon, Saskatchewan. The steers were purchased over 5 days in 2005 (Table 3.1) in order to allow them to be processed on the day they were purchased. The steers were transported to the University of Saskatchewan Beef Cattle Research Unit, where they underwent processing which included: implantation with Zeranol (an estrogen analog), prophylactic oxytetracyline administration, vaccination against clostridial disease, application of Ivomec pour-on for treatment of external parasites, ear tagging and weighing. Cattle purchased and processed on one day were housed together in order to ensure that the behavioural testing could be performed at precise intervals.

Table 3.1 A timeline for all procedures performed on the cattle during the course of this study

Date	
November 29, 2005	Purchased/Processed (Group A)
November 30, 2005	Behaviour measurement session 1 (A) & Purchased/Processed (Group B)
December 1, 2005	Behaviour measurement session 1 (B) & Purchased/Processed (Group C)
December 2, 2005	Behaviour measurement session 1 (C)
December 6, 2005	Purchased/Processed (Group D)
December 7, 2005	Behaviour measurement session 1 (D) & Purchased/Processed (Group E)
December 8, 2005	Behaviour measurement session 1 (E)
January 4, 2006	Blood Collected & Weighed (A)
January 5, 2006	Blood Collected & Weighed (B)
January 9, 2006	Blood Collected & Weighed (C)
January 10, 2006	Blood Collected & Weighed (D)
January 12, 2006	Blood Collected & Weighed (E)
January 24, 2006	Behaviour measurement session 2 (A)
January 25, 2006	Behaviour measurement session 2 (B)
January 26, 2006	Behaviour measurement session 2 (C)
January 31, 2006	Behaviour measurement session 2 (D)
February 1, 2006	Behaviour measurement session 2 (E)
February 7, 2006	Weighed
March 21, 2006	Behaviour measurement session 3 (A)
March 22, 2006	Behaviour measurement session 3 (B)
March 23, 2006	Behaviour measurement session 3 (C)
March 28, 2006	Behaviour measurement session 3 (D)
March 29, 2006	Behaviour measurement session 3 (E)
March 31, 2006	Weighed (End of backgrounding weight)
April 5&6, 2006	Ultrasound backfat and Rib-eye Area measured
May 2, 2006	Weighed (Start of finishing weight)
May 3, 2006	Shipped to finishing facility
July 31, 2006	End of finishing weight
August 1, 2006	End of finishing weight
September 5, 2006	42 steers slaughtered
September 6, 2006	42 steers slaughtered
September 7, 2006	42 steers slaughtered
September 8, 2006	42 steers slaughtered
September 11, 2006	46 steers slaughtered
September 12, 2006	46 steers slaughtered
September 13, 2006	43 steers slaughtered
September 14, 2006	43 steers slaughtered
September 15, 2006	43 steers slaughtered

3.2 Behaviour measurements

A series of behaviour measurements were performed in order to assess each animal's temperament (Figure 3.1). The order of testing for each individual was: Subjective Score (SS), Strain Gauge (SG), Movement Measurement Device (MMD) and Exit Time (ET). The entire series of measurements were repeated 3 times at two-month intervals; the first measurement session was conducted for each pen the day after processing (Table 3.1). The number of the measurement session follows the trait being discussed, *e.g.* SS 1 refers to the subjective score from session one.

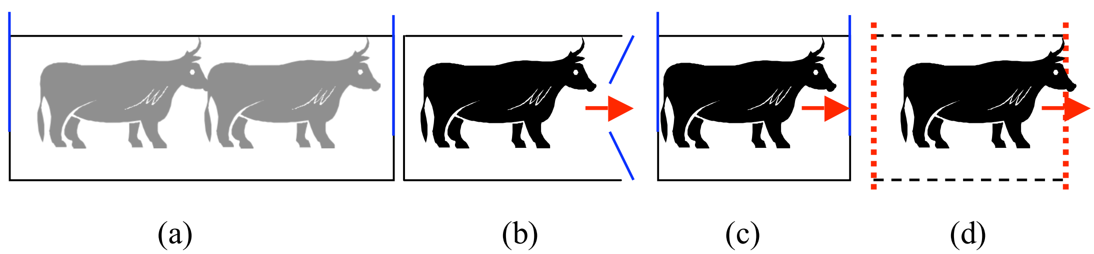


Figure 3.1 A schematic representation of the temperament measurement sequence. Portion (a) represents the two steers waiting behind the subject steer, which is shown in black. Section (b) represents the squeeze chute where Subjective Score and Strain Gauge Absolute Strain Force were measured, (c) represents the scale and Movement Measurement Device, and (d) represents the section of panels where Exit Time was measured, with the light beams as dotted red lines. Section (a), (b) and (c) were indoors, while (d) was outdoors.

The pens of steers were measured in the same order each time so that a consistent 2 month interval between measurement sessions was maintained (Table 3.1). During each measurement session, great care was taken to handle the steers calmly and with as little noise as possible in order to minimize the influence of individual handlers or handling styles on the steers' reactions. We also took care to ensure that all tables etc. in the handling facility were identically located during each session so that novel objects in the facility were not influencing the animals' behaviour. For similar reasons, we also ensured that all observers that were visible to the cattle were the same for each session. During the measurement sessions, there were three steers in the handling facility at all times – one being measured, and two waiting behind in the chute (Figure 3.1). For the last pen of the day, the first two animals measured were put through the chute again to serve as the "following" animals for the last steer.

3.2.1 Subjective Score

A SS was given to each steer while they were restrained in the head gate. The 1-5 scale (Table 3.2) used for the SS was based on one used by Grandin (1993). Scoring began 5 seconds after the head gate was closed, in order to allow the animals to settle somewhat after the shock of being caught in the gate. Observation continued for a 10 second period. During the first five seconds of this observation period, the head gate operator stood quietly in the steer's view, while during the last five seconds she gently held the steer's ear. In order to minimize inter-observer differences in scoring, a single observer performed all of the subjective scoring.

Table 3.2 The 1-5 scale used for allocation of a Subjective Score to each steer (adapted from Grandin 1993)

Subjective Score	Description
1	Little or no movement
2	Low amplitude movements, < 3 violent kicks, head shakes or rears
3	High amplitude movements, ≥ 3 violent kicks, head shakes or rears
4	Nearly continuous violent kicking, head shaking or rearing
5	Continuous, extremely violent movements (no pauses)

3.2.2 Strain Gauge

The SG is a device for quantifying an animal's response to restraint, and was previously used to evaluate response to the pain of branding (Schwartzkopf-Geshwein *et al.* 1997). The head gate of a squeeze chute (Figure 3.2) was used to mount the tiny strain gauges (Figure 3.3). The strain gauges measure the amount of force (mV) a steer applies to the head gate while restrained, at a sampling rate of 20 readings per sec. for 10 sec., for a total of 200 data points. These data are sent to a data logger, and subsequently saved as a data file, with one file per subject.

Figure 3.4 illustrates the nature of the raw SG data. The raw SG data contains both positive and negative values (Figure 3.4) since the steers both pulled and pushed on the head gate. The SG is calibrated such that the neutral position of the gate gives a zero mV value, a steer pulling backwards on the gate gives positive values, and a steer pushing forward on the gate gives negative values. In order to account equally for both the pulling and pushing motions, we used the sum of the absolute values of the 200 mV data points (Absolute Strain Force, ASF) as the objective measurement that was compared between steers.

The 10 seconds of SG data were collected concurrently with the SS, following the same 5 sec. recovery period after the steer was caught in the head gate. The SG began recording data before the steer entered the head gate, and continued recording after the steer was released. Therefore, an assistant carefully recorded the exact beginning and end of the measurement period within each data file.

Figure 3.2 A picture of the head gate to which the strain gauge device was attached

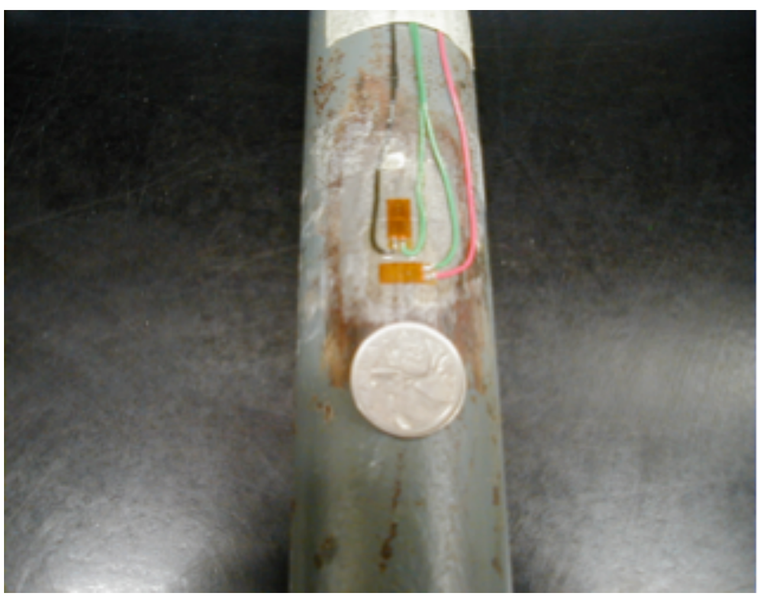
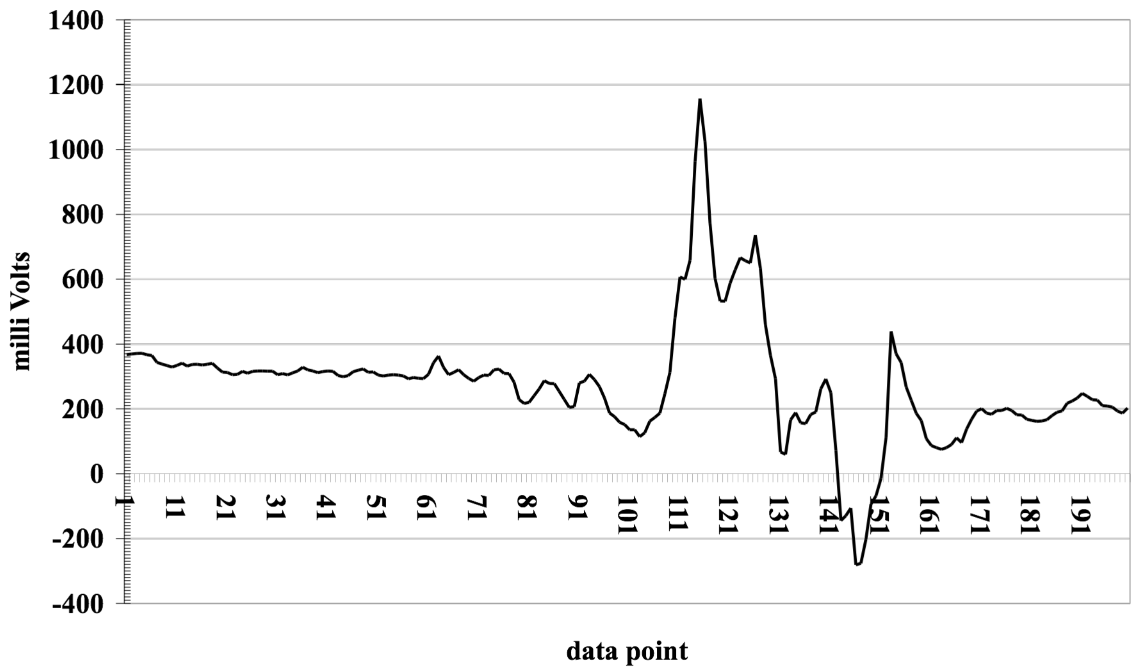


Figure 3.3 A picture of the strain gauges in place on the bar of the head gate, shown with a quarter for scale



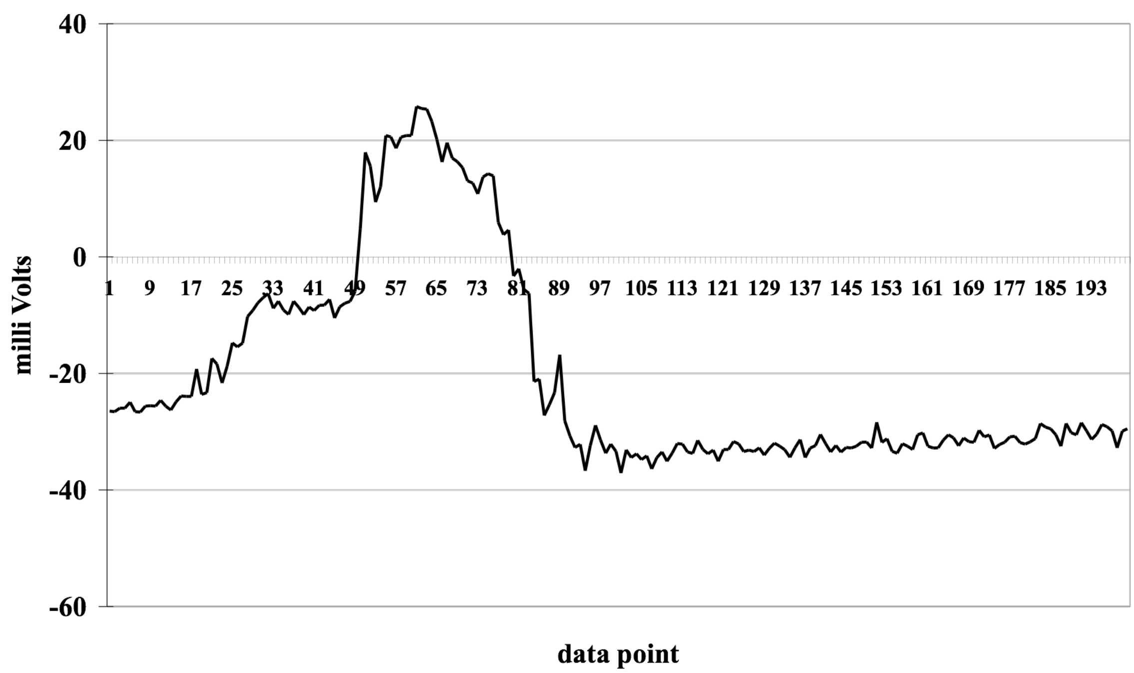


Figure 3.4 A graphical representation of the raw Strain Gauge data from two steers, showing the variation in milliVolts of strain over the 200 data points

3.2.3 Movement Measurement Device

Stookey *et al.* (1994) developed the MMD as a tool for objectively quantifying cattle response to isolation and confinement. The device attaches to the load cells of a scale, and records the fluctuations in voltage created by the animal's movements, at a rate of 20 readings per sec. for a period of one minute. The MMD records a "peak" when a trend of increasing or decreasing voltage reverses, and also calculates the standard deviation of theses peaks and the mean voltage, with the mean corresponding to the animal's weight. After the one minute recording period, the number of peaks, standard deviation and mean are displayed on a readout screen and then carefully recorded by an observer. For this trial we chose to analyze only the number of MMD peaks, as we felt that this measurement was the most intuitively understood of the three. The MMD testing was conducted in a closed sided weight scale. The steers entered this after traveling a short distance upon release from the head gate where the SS and SG observations were conducted (Figure 3.1).

3.2.4 Exit Time

The ET test was conducted immediately after the cattle left the MMD weigh scale (Figure 3.1). The steers' ET was measured by an optical timing device which recorded the time in sec. it took the steer to break two infra red light beams placed 2.9 m apart (Figure 3.5). We did not convert the times to flight speed *i.e.* we used ET as seconds rather than m/sec.

Figure 3.5 The metal panels used to form the Exit Time test chute. The yellow circles indicate the position of the light beams for the optical timing device.

3.2.5 Habituation

Habituation was calculated according to Schmutz *et al.* (2001), simply as the difference in the various measurements between the first and last measurement sessions. For example, habituation for number of MMD peaks for any one steer is (number of MMD peaks in session 3) – (number of MMD peaks in session 1); the resulting value was then compared between animals.

3.3 Blood Collection & DNA extraction

Blood was collected from each steer by jugular venipuncture into vaccutainers containing EDTA one month after purchase (Table 3.1). Genomic DNA was extracted from the collected blood using a salt extraction method (Montgomery & Sise, 1990).

3.4 PCR-RFLP

Although we started our study with 400 steers, two of them died before we were able to get blood samples from them, therefore 398 animals were genotyped for each SNP. Genotyping was carried out using the previously reported Polymerase Chain Reaction-Restriction Fragment Length Polymorphism (PCR-RFLP) tests for the *LEP 73C>T* (Buchanan *et al.* 2002a) and *CRH 22C>G* (Buchanan *et al.* 2005) SNPs. For the third SNP, *CRH 240C>G*, the PCR-RFLP primers and protocol used were slightly modified from those reported by Buchanan *et al.* (2002b). The PCR-RFLP protocols are detailed in the following three sections.

3.4.1 Leptin *73C>T*

The *LEP 73C>T* SNP was amplified in a 20 μl cocktail, containing: 4 pmol each of the forward and reverse primer, 0.2 mM dNTPs, 2.0 μl Invitrogen 10X PCR Buffer, 1.5 mM $MgCl_2$, 0.5 U *Taq* DNA polymerase (Invitrogen), 14.5 μl of sterile ddH_2O and 100 ng of genomic DNA template.

The PCR amplification program consisted of an initial 2 min denaturing phase at 94 °C, followed by 35 cycles of denaturing for 45 sec at 94 °C, annealing for 45 sec at 60 °C, and extension for 55 sec at 72 °C, with a final 3 min extension phase at 72 °C.

The 25 μl restriction endonuclease digestion consisted of 2 μl Y+/Tango Buffer (MBI Fermentas), 2 μl sterile ddH_2O, and 1 μl of *Kpn21* (10 U/μl, MBI Fermentas) was added to the 20 μl PCR product and digested in a 55 °C waterbath for 2 hours. The resulting DNA fragments were separated and visualized on a 3% agarose gel. Digestion of the 94 bp PCR product results in the C allele cutting into 75 and 19 bp fragments, while the T allele remains uncut.

3.4.2 CRH 22C>G

The *CRH 22C>G* SNP was amplified in a 25 μl cocktail, containing: 4 pmol each of the forward and reverse primer, 0.16 mM dNTPs, 2.0 μl Jeffrey's Buffer (JB, contains 45 mM Tris-HCl, 11 mM $(NH_4)_2SO_4$, 4.5 mM $MgCl_2$, 6.7mM β-mercaptoethanol, 4.5 mM EDTA and 0.25 mM Spermidine) 2.0 μl Dimethyl sulfoxide (10% DMSO), 0.5 U DNA polymerase (Invitrogen), 18.1 μl of sterile ddH_2O and 100 ng of genomic DNA template.

The PCR amplification program consisted of an initial 2 min denaturing phase at 94 °C, followed by 39 cycles of denaturing for 1 min at 94 °C, annealing for 45 sec at 60 °C, and extension for 50 sec at 72 °C, with a final 4 min extension phase at 72 °C.

The 30 μl restriction endonuclease digestion consisted of 3 μl NE Buffer 3 (New England Biolabs), 1 μl sterile ddH2O, and 1 μl of *DdeI* (10 U/μl) added to the 25 μl PCR product and digested in a 37 °C waterbath for 4 hours. The resulting DNA fragments were separated and visualized on a 4% agarose gel. Digestion of the 129 bp PCR product results in all samples cutting into a 41 bp fragment plus either a 19 and a 69 bp fragment (G allele) or an 88 bp fragment (C allele).

A previous graduate student performed the genotyping for the *CRH 22C>G* SNP. In order to verify this work, all of the gels were re-examined with the new observer blind to the original genotype calls. Any samples for which the genotype calls disagreed between observers, or where the original gel was somewhat unclear, were re-tested. Numerous randomly selected samples were also re-tested to confirm that the genotyping results were identical between testers.

3.4.3 CRH 240C>G

The *CRH 240C>G* SNP was amplified in a 20 µl cocktail, containing: 4 pmol each of the forward and reverse primer (forward 5'-TGC CCG GCA GGC ATC ACA G-3'; reverse 5'-AGA GAG GGG AGC AGC CCG-3'), 0.2 mM dNTPs, 2.0 µl Jeffrey's Buffer (JB), 1.3 mM Betaine, 0.5 U *Taq* DNA polymerase (Qiagen), 9.9 µl of sterile ddH_2O and 100 ng of genomic DNA template.

The PCR amplification program consisted of an initial 2 min denaturing phase at 94 °C, followed by 35 cycles of denaturing for 1 min at 94 °C, annealing for 50 sec at 66 °C, and extension for 50 sec at 72 °C, with a final 4 min extension phase at 72 °C.

The 25 µl restriction endonuclease digestion consisted of 2 µl Taq1+ 10X Buffer (Qiagen), 1 µl sterile ddH_2O, 1 µl spermidine (80 mM) and 1 µl of *Taq1* (10 U/µl, MBI Fermentas) restriction endonuclease, added to the 20 µl PCR product and digested in a 65 °C waterbath for 2.5 hours. The resulting DNA fragments were separated and visualized on a 2.5% agarose gel. The digestion of the 157 bp PCR product results in the G allele cutting into 130 and 27 bp fragments, while the C allele remains uncut.

3.5 Growth & Carcass measurements

3.5.1 Backgrounding

The steers were fed a backgrounding diet (Appendix A) for an average of 118 days, with a range of 114 to 122 days, depending on purchase lot. Initial weights were collected for each steer at the time of processing, and additional weights were taken at the time of blood collection, on Feb. 7th, March 31st and May 2nd, 2006 (Table 3.1). March 31st was considered the end of backgrounding (EOB) weight, while May 2nd was just prior to the steers' shipment to Pound-Maker and was therefore recorded as the steer's start of finishing weight. Backgrounding average daily gain (ADG) was calculated using the end of backgrounding weight. Ultrasound was used to measure rib-eye area (REA) and backfat cover between the 12th and 13th ribs on April 5th and 6th.

3.5.2 Finishing & Carcass Data

The steers were shipped to Pound-Maker feedlot on May 3rd, 2006, where they were housed in two pens of ~200 animals, and all were fed a standard finishing diet (Appendix B). The end of finishing weights used to calculate finishing ADG were taken on July 31st and Aug. 1st, 2006 at 119 and 120 days on feed, respectively. Finishing ADG was calculated using the difference between the start and end of finishing weights, less 4% for shrink, divided by 119.5 days on feed.

The steers were shipped to XL Beef in Moose Jaw, SK. for slaughter in 9 lots (Table 3.1). The small slaughter lots of ~ 45 animals per day were necessary in order to enable a carcass grader to record Blue Tag data for all of the carcasses. The Blue Tag data for each carcass included warm carcass weight, average fat, grade fat, REA, marbling, quality grade and cutability.

3.6 Statistical analysis

Initial statistical analysis was performed using the program Statview to run simple regressions between the genotypes at each SNP and the various behaviour and growth/carcass measurements.

For the habituation data, a paired t-test was performed to determine if mean values for all behaviour measurements were significantly different between sessions 1 and 3. Additionaly, the univariate procedure of SAS 9.1 (2003) was used to test the behaviour measurements for normality.

The mixed procedure of SAS 9.1 (2003) was then used to analyze the effects of the *LEP*, *CRH 22C>G* and *CRH 240C>G* SNPs on the behaviour, growth and carcass traits. Initial analysis for each behaviour measurement was performed by including all three SNP genotypes, as well as two way interactions between the *LEP* and *CRH 22C>G* and between the *LEP* and *CRH 240C>G* genotypes in the models. The general equation for the initial model was:

$$Y = \mu + LEP + CRH\ 22C{>}G + CRH\ 240C{>}G + LEP \times CRH\ 22C{>}G + LEP \times CRH\ 240C{>}G + e$$

Further analysis was performed by removing the non-significant (P>0.05) effects from the models for the above mentioned traits.

For the ASF values, an initial analysis was preformed including each animal's estimated weight on the measurement day (initial weight + (ADG x days since processing)) as a main effect because we initially hypothesized that an animal's weight should have an effect on the ASF values. Estimated weight had no significant effect on ASF, therefore it was removed from the models during further analyses. We also added start of test weight into the preliminary model as a covariate to determine if it had an effect on any of the other behaviour measurements. Start of test weight was not a significant effect, therefore it was not included in any further analyses.

4 RESULTS

4.1 PCR-RFLP

Figures 4.1, 4.2 and 4.3 show representative agarose gels for the *LEP*, *CRH 22C>G* and *CRH 240C>G* PCR-RFLPs.

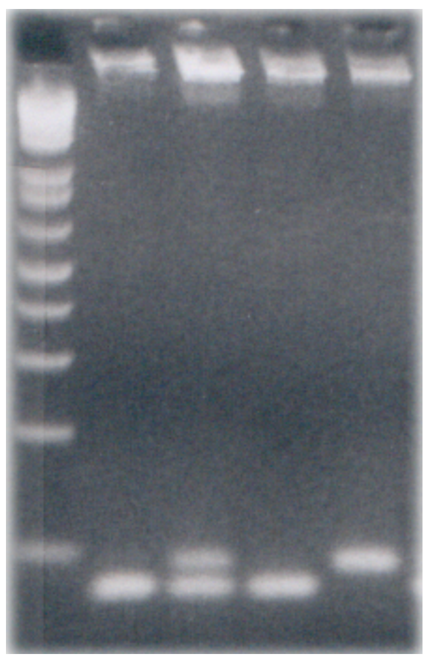


Figure 4.1 A representative gel for the *LEP* PCR-RFLP, with the 1 kb+ ladder in lane 1, and *CT*, *CC* and *TT* steers in lanes 3, 4, and 5 respectively

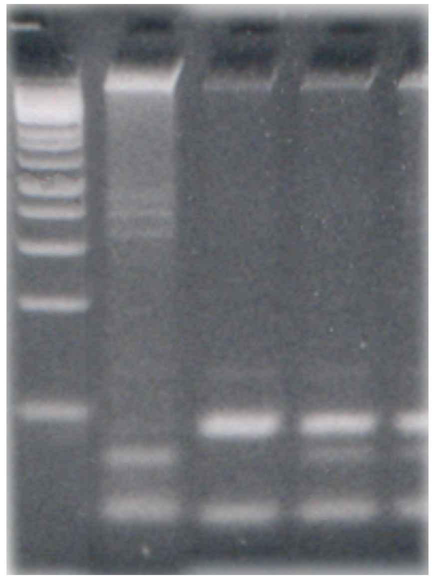


Figure 4.2 A representative gel for the *CRH 22C>G* PCR-RFLP, with the 1 kb+ ladder in lane 1, and *GG*, *CC* and *CG* steers in lanes 2, 3 and 4 respectively

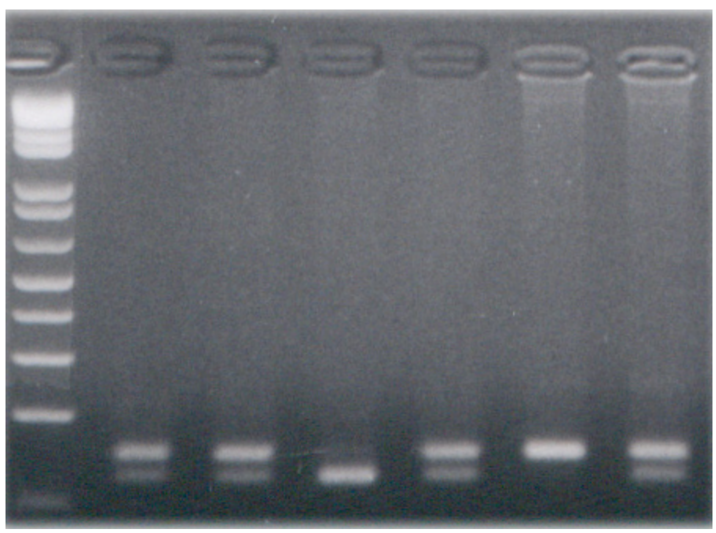


Figure 4.3 A representative gel for the *CRH 240C>G* PCR-RFLP, with the 1 kb+ ladder in lane 1, and *GG*, *CG* and *CC* steers in lanes 4, 5, and 6, respectively

The genotyping results and allele frequencies for the *LEP*, *CRH 22C>G* and *CRH 240C>G* SNPs are presented in Table 4.1(see Appendix C for complete genotype data).

Table 4.1 Number of steers of each SNP genotype and allele frequencies for each SNP

	LEP		***CRH 22C>G***		***CRH 240C>G***	
Number per genotype:	CC	97	CC	92	CC	31
	CT	210	CG	178	CG	152
	TT	91	GG	128	GG	215
Allele Frequency:	C	0.508	C	0.455	C	0.269
	T	0.492	G	0.545	G	0.731

4.2 Descriptive Statistics

4.2.1 Behaviour Measurements

Behaviour data were collected for all three measurement sessions, however only data from sessions one and three were analyzed in order to assess temperament and habituation. The first session is of interest as it reflects each animal's temperament while they are relatively naïve to the facilities and handling in general. The measurements from the 3rd session reflect the animals' temperament after a period of learning and acclimation. The second measurements were conducted essentially as an attempt to induce habituation, and are not considered to be as informative as the first and last sessions. A summary of the results from sessions 1 and 3 are presented (see Appendix D for complete behaviour data). The average SS for both session 1 and session 3 was approximately 2 () on the 1-5 rating scale (Table 3.2), and less than 2% of the steers received a SS of 4 or 5. The univariate procedure of SAS (9.1) showed that the behaviour data were not normaly distributed (Table 4.3). There were no differences between the strain gauge values when the steers' ears were being handled or not, and therefore the entire 10 second period was considered (Sebastian *et al.* 2007).

Table 4.2 Summary of the behaviour measurements from the first and third measurement sessions

	SS 1	ASF 1	MMD Peak 1	ET 1 (sec)
Avg.	1.91	91966.51	22.51	3.48
SD	0.675	58319.410	24.702	1.420
Min.	1	3928.96	0	0.98
Max.	5	569319.24	147	14.49
n	399	396	400	400

	SS 3	ASF 3	MMD Peak 3	ET 3 (sec)
Avg.	2.12	90411.48	61.51	4.66
SD	0.557	58837.252	47.679	6.430
Min.	1	2230.4	2	0.80
Max.	5	348595	286	62.70
n	397	393	398	398

SS=Subjective Score, ASF=Absolute Strain Force,
MMD=Movement Measurement Device, ET=Exit Time

Table 4.3 Tests for normality results for the behaviour measurements

Behaviour Measurement	Shapiro-Wilk P Value	Skewness	Kurtosis
SS 1	<0.0001	0.5883	1.4343
ASF 1	<0.0001	2.0175	11.3248
MMD Peaks 1	<0.0001	1.9054	3.7080
ET 1	<0.0001	3.0756	18.6264
SS 3	<0.0001	0.9269	3.3621
ASF 3	<0.0001	1.3843	2.8480
MMD Peaks 3	<0.0001	1.3446	2.2611
ET 3	<0.0001	5.5198	36.2840
SS 3-1	<0.0001	-0.8750	5.9504
ASF 3-1	<0.0001	-0.9835	11.3071
MMD Peaks 3-1	<0.0001	0.9025	0.9434
ET 3-1	<0.0001	5.3851	36.1750

SS=Subjective Score, ASF=Absolute Strain Force,

MMD=Movement Measurement Device, ET=Exit Time

4.2.2 Habituation

We expected, based on previous work, that the steers would grow accustomed to handling and as a result their temperament would improve between measurement 1 and measurement 3 (Schmutz *et al.* 2001). In order to evaluate habituation, the various scores from measurement 1 are subtracted from those of measurement 3. Negative values for SS, MMD Peak and ASF indicated that the parameter improved, while positive values for ET indicated improvement.

From the numerical data in Table 4.4, it seems that habituation did not take place (i.e. the average scores did not improve over the course of the three measurement sessions) for SS or MMD Peak, while habituation did take place for ASF and ET. The results of the paired t-tests show that the averages for SS 1 & 3, , MMD Peak 1 & 3, and ET 1 & 3 are significantly different ($P<0.05$,Table 4.5). The change in the average of each behaviour measurement can be visualized in Figures 4.4 through 4.7.

Table 4.4 Summary of habituation data for the behaviour measurements. Negative values for SS, MMD Peak and ASF indicate that the parameter improved, while improvement is shown by positive values for ET.

	SS 3-1	ASF 3–1	MMD Peak 3-1	ET 3-1
Avg.	0.199	-666.21	39.03	1.169
SD	0.7339	68703.480	40.483	6.383
Min	-5	-541120.24	-59	-11.62
Max	2	332894.34	194	59.85
n	396	389	398	397

SS=Subjective Score, ASF=Absolute Strain Force, MMD=Movement Measurement Device, ET=Exit Time

Table 4.5 Paired t-test results for the differences between means for each measurement between sessions 1 and 3

Behaviour Measurement		P Value
Subjective Score 1	Subjective Score 3	<0.0001
Absolute Strain Force 1	Absolute Strain Force 3	0.7431
MMD Peaks 1	MMD Peaks 3	<0.0001
Exit Time 1	Exit Time 3	0.0006

MMD = Movement Measurement Device

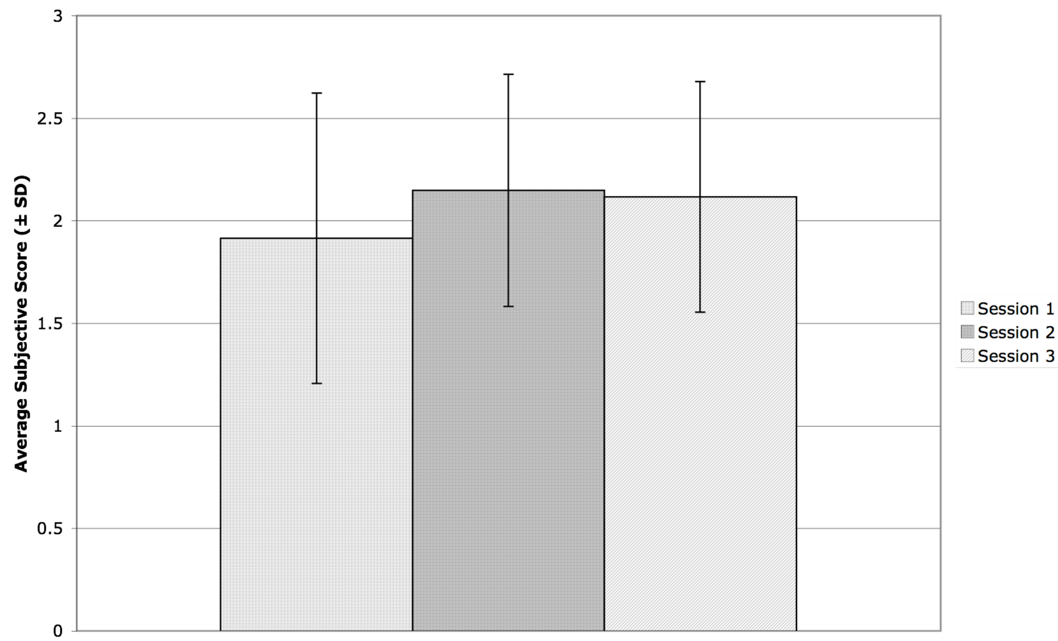


Figure 4.4 Average Subjective Score (± SD) for each measurement session

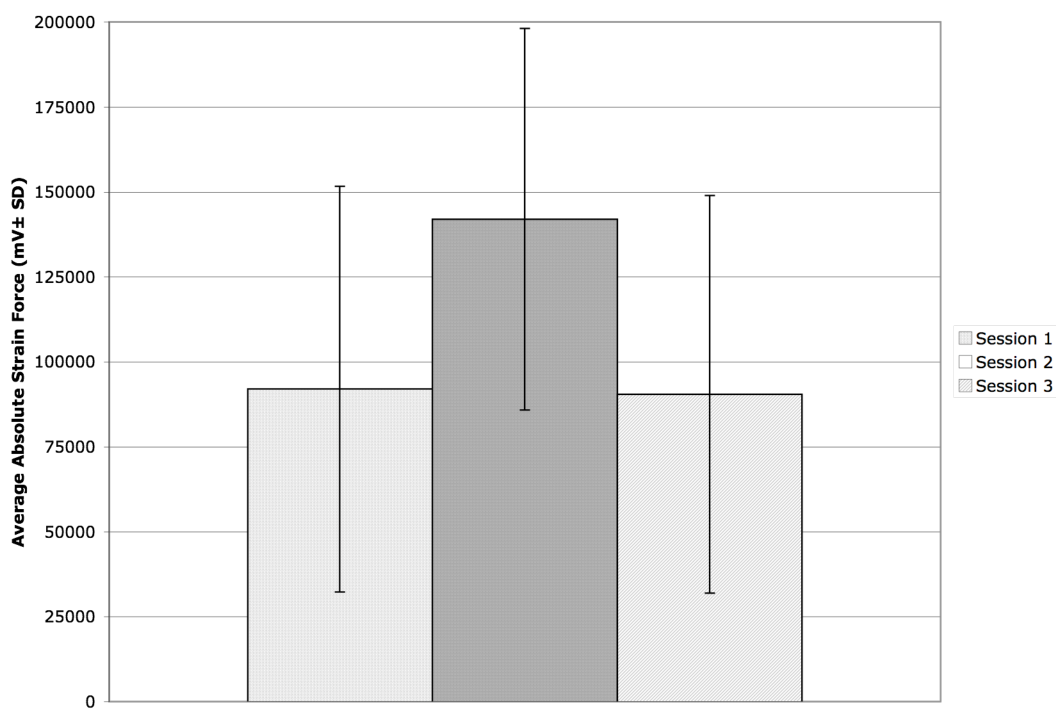


Figure 4.5 Average Absolute Strain Force (± SD) for each measurement session

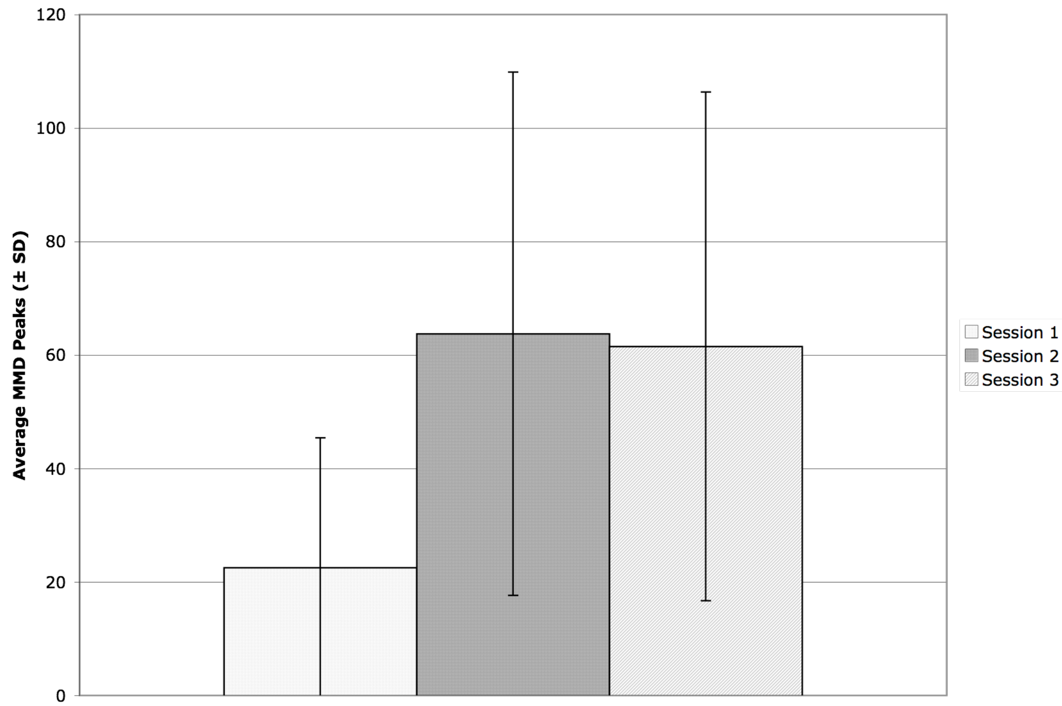


Figure 4.6 Average number of MMD Peaks (± SD) for each measurement session

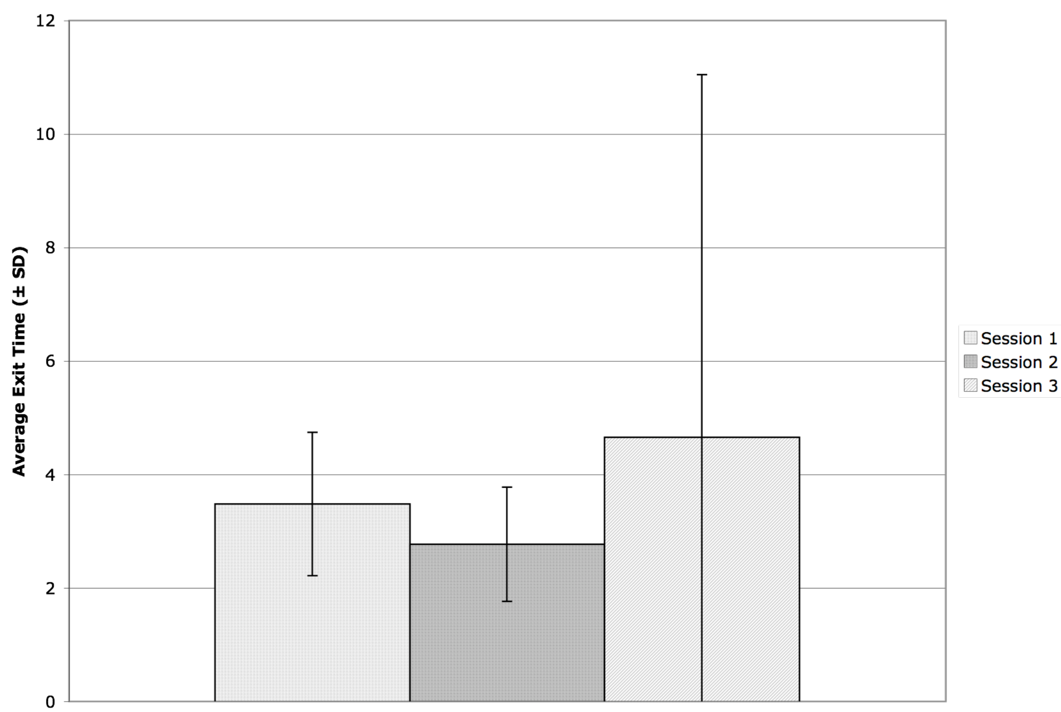


Figure 4.7 Average Exit Time (± SD) for each measurement session

4.2.3 Growth & Carcass measurements

A summary of the steers' weight and ADG at the end of backgrounding (EOB), as well as the ultrasound backfat (BF) and REA measurements, is presented in Table 4.6 (see Appendix E for complete data).

At the time of slaughter, some of the steers had missing or incorrect ear tags, therefore only carcass data for which we could unequivocally identify the steer's original ID number are presented. A summary of the steers' weight and ADG after finishing at Pound-Maker feedlot, as well as Blue Tag carcass data, is presented in Table 4.5. The number of carcasses of each Quality Grade is presented in Table 4.8 (see Appendix F for complete data).

Table 4.6 Summary of growth and carcass measurements after backgrounding

	Start of Test	End of Backgrounding		Ultrasound	
	Weight (kg)	Weight (kg)	ADG (kg/day)	Backfat (mm)	REA (cm^2)
Avg.	243.66	385.24	1.20	2.25	64.53
SD	19.41	30.26	0.178	1.352	7.570
Min	187.5	293	0.72	0	47.05
Max	308	482	1.84	8.20	89.66
N	400	398	398	392	392

ADG=Average Daily Gain, REA=Rib-eye Area

Table 4.7 Summary of growth and carcass measurements after finishing

	End of finishing Wt (kg)	ADG (kg/day)	WC Wt (kg)	Avg. Fat	Grade Fat	REA (cm^2)	Marbling	Cutability (%)
Avg.	631.70	1.64	384.68	9.75	8.31	101.58	7.71	61.08
SD	53.817	0.239	153.84	3.640	3.610	11.763	0.559	3.100
Min	478.54	0.89	294.80	2	1	76	7	50
Max	784.71	2.31	462.40	24	23	131	9	65
n	384	384	386	386	386	386	377	386

WT=Weight, ADG=Average Daily Gain, WC=Warm Carcass, REA=Rib-eye Area

Table 4.8 The number (and %) of carcasses in each Quality Grade

Quality Grade	n	%
AAA	130	33.7
AA	227	58.9
A	20	5.2
B4	9	2.3

4.3 Statistical Analysis

4.3.1 Simple Regressions

Initial statistical analysis was performed using the program Statview to run simple regressions between the genotypes at each SNP and the various behaviour measurements and the growth/carcass measurements. The genotypes at each SNP were converted into the number of one allele present for analysis purposes *e.g. LEP* CC, CT and TT was represented as 0, 1 and 2 copies of the T allele respectively. Simple regression results for the genotypes and behaviour measurements are presented in Table 4.9 (*LEP*), Table 4.10 (*CRH 22C>G*) and Table 4.11 (*CRH 240C>G*). The results for the genotypes and the growth and carcass measurements are presented in Table 4.12 (*LEP*), Table 4.13 (*CRH 22C>G*) and Table 4.14 (*CRH 240C>G*).

Table 4.9 Results of the simple regression analysis between the number of LEP T alleles and the behaviour measurements

	LEP – # of T alleles			
Behaviour	R	R^2	P	+/-
SS 1	0.046	0.002	0.3604	
MMD Peak 1	**0.137**	**0.019**	**0.0062**	**-**
ET 1	0.036	0.001	0.4758	
ASF 1	0.031	0.001	0.5417	
SS 3	0.009	<0.0001	0.8645	
MMD Peak 3	**0.118**	**0.014**	**0.0182**	**-**
ET 3	0.058	0.003	0.2524	
ASF 3	0.022	<0.0001	0.6672	
SS 3-1	0.043	0.002	0.3912	
MMD Peak 3-1	0.056	0.003	0.2677	
ASF 3-1	0.011	<0.0001	0.8312	
ET 3-1	0.066	0.004	0.1895	

Bold (P<0.05), *Italic (P<0.10)*

SS=Subjective Score, ASF=Absolute Strain Force, MMD=Movement Measurement Device, ET=Exit Time

+/- indicates the slope of the regression line for significant results

Table 4.10 Results of the simple regression analysis between the number of *CRH 22C>G* G alleles and the behaviour measurements

Behaviour	R	R^2	P	+/-
	CRH 22C>G - # of G alleles			
SS 1	0.056	0.003	0.2685	
MMD Peak 1	0.033	0.001	0.5084	
ET 1	*0.083*	*0.007*	*0.0987*	+
ASF 1	0.008	<0.0001	0.8706	
SS 3	0.018	<0.0001	0.7205	
MMD Peak 3	0.019	<0.0001	0.7027	
ET 3	0.050	0.003	0.3195	
ASF 3	0.056	0.003	0.2638	
SS 3-1	0.074	0.005	0.1440	
MMD Peak 3-1	0.043	0.002	0.3932	
ASF 3-1	0.067	0.004	0.1905	
ET 3-1	0.067	0.004	0.1905	

Bold (P<0.05), *Italic (P<0.10)*

SS=Subjective Score, ASF=Absolute Strain Force,
MMD=Movement Measurement Device, ET=Exit Time

+/- indicates the slope of the regression line for significant results

Table 4.11 Results of the simple regression analysis between the number of *CRH 240C>G* G alleles and the behaviour measurements

Behaviour	R	R^2	P	+/-
	CRH 240C>G - # of G alleles			
SS 1	0.040	0.002	0.4283	
MMD Peak 1	**0.102**	**0.010**	**0.0420**	**-**
ET 1	0.066	0.004	0.1904	
ASF 1	0.040	0.002	0.4333	
SS 3	*0.088*	*0.008*	*0.0808*	-
MMD Peak 3	0.064	0.004	0.1991	
ET 3	*0.085*	*0.007*	*0.0896*	-
ASF 3	0.034	0.001	0.5065	
SS 3-1	0.034	0.001	0.5041	
MMD Peak 3-1	0.014	<0.0001	0.7862	
ASF 3-1	0.051	0.003	0.3128	
ET 3-1	**0.101**	**0.010**	**0.0446**	**-**

Bold (P<0.05), *Italic (P<0.10)*

SS=Subjective Score, ASF=Absolute Strain Force,
MMD=Movement Measurement Device, ET=Exit Time

+/- indicates the slope of the regression line for significant results

Table 4.12 Results of the simple regression analysis between the number of *LEP* T alleles and the growth and carcass measurements

	LEP – # of T alleles			
Measurement	R	R^2	P	+/-
SOT Wt	*0.090*	*0.008*	*0.0766*	+
EOB Wt	0.019	<0.0001	0.7035	
EOB ADG	0.017	<0.0001	0.7458	
EOB BF	**0.106**	**0.011**	**0.0376**	+
EOB REA	0.037	0.001	0.4764	
End of finishing Wt	0.048	0.002	0.3496	
Finishing ADG	0.074	0.005	0.1505	
WC Wt	0.069	0.005	0.1743	
Avg. Fat	**0.173**	**0.030**	**0.0007**	+
Grade Fat	**0.170**	**0.029**	**0.0008**	+
REA	0.008	<0.0001	0.8696	
Cutability	**0.163**	**0.026**	**0.0015**	**-**
Marbling	0.048	0.002	0.3508	

Bold (P<0.05), *Italic (P<0.10)*

SOT=Start of Test, WT=Weight, EOB=End of Backgrounding, ADG=Average Daily Gain, BF=Backfat, REA=Rib-eye Area, WC=Warm Carcass
+/- indicates the slope of the regression line for significant results

Table 4.13 Results of the simple regression analysis between the number of *CRH 22C>G* G alleles and the growth and carcass measurements

	CRH 22C>G – # of G alleles			
Measurement	R	R^2	P	+/-
SOT Wt	<0.0001	<0.0001	0.9956	
EOB Wt	0.012	<0.0001	0.8095	
EOB ADG	0.043	0.002	0.4004	
EOB BF	0.031	0.001	0.5497	
EOB REA	0.079	0.006	0.1220	
End of finishing Wt	0.020	<0.0001	0.6953	
Fin. ADG	0.021	<0.0001	0.6774	
WC Wt	0.031	0.001	0.5393	
Avg. Fat	<0.0001	<0.0001	0.9945	
Grade Fat	0.008	<0.0001	0.8731	
REA	0.035	0.001	0.4882	
Cutability	0.019	<0.0001	0.7191	
Marbling	*0.092*	*0.009*	*0.0729*	+

Bold (P<0.05), *Italic (P<0.10)*

SOT=Start of Test, WT=Weight, EOB=End of Backgrounding, ADG=Average Daily Gain, BF=Backfat, REA=Rib-eye Area, WC=Warm Carcass

+/- indicates the slope of the regression line for significant results

Table 4.14 Results of the simple regression analysis between the number of *CRH 240C>G* G alleles and the growth and carcass measurements

	CRH 240C>G – # of G alleles			
Measurement	R	R^2	P	+/-
SOT Wt	0.038	0.001	0.4516	
EOB Wt	0.074	0.005	0.1488	
EOB ADG	0.054	0.003	0.2865	
EOB BF	*0.099*	*0.010*	*0.0540*	+
EOB REA	*0.097*	*0.009*	*0.0590*	-
End of finishing Wt	0.046	0.002	0.3697	
Fin. ADG	0.014	<0.0001	0.7814	
WC Wt	0.001	<0.0001	0.9785	
Avg. Fat	*0.091*	*0.008*	*0.0738*	+
Grade Fat	*0.099*	*0.010*	*0.0510*	+
REA	**0.105**	**0.011**	**0.0391**	**-**
Cutability	*0.087*	*0.008*	*0.0917*	-
Marbling	0.006	<0.0001	0.9042	

Bold (P<0.05), *Italic (P<0.10)*

SOT=Start of Test, WT=Weight, EOB=End of Backgrounding, ADG=Average Daily Gain, BF=Backfat, REA=Rib-eye Area, WC=Warm Carcass

+/- indicates the slope of the regression line for significant results

4.3.2 Mixed Model ANOVA: Behaviour and genotype

Significant main effects of genotype were observed for some traits (Table 4.15) therefore following the preliminary ANOVA analysis, the models where a significant genotype effect was found were simplified by removing non-significant variables (P>0.05) in order to obtain final LSM and P values.

Table 4.15 A summary of the preliminary Tests of Main Effects for SNP genotypes on behaviour

Behaviour	Test of Main Effects – P values				
	LEP	*CRH 22C>G*	*CRH 240C>G*	*LEP* x *CRH 22C>G*	*LEP* x *CRH 240C>G*
SS 1	0.5879	0.9679	0.6339	0.6181	0.4239
ASF 1	**0.0391**	0.9487	0.5237	**0.0396**	**0.0158**
MMD Peak 1	0.1349	0.4538	**0.0262**	0.9088	0.7268
ET 1	0.6499	0.6998	0.9229	0.9550	0.9148
SS 3	0.9500	0.1194	**0.0209**	0.6785	0.8667
ASF 3	0.4281	*0.0780*	0.2475	0.2978	0.8256
MMD Peak 3	0.1632	0.5468	*0.0619*	0.4010	0.7274
ET 3	**0.0014**	0.9085	**0.0470**	0.6267	**0.0014**
SS 3-1	0.5147	0.1945	0.3169	0.2078	0.2899
ASF 3-1	0.2922	*0.0660*	0.1287	0.2102	*0.0911*
MMD Peak 3-1	0.3480	0.5007	0.5280	0.3802	0.8187
ET 3-1	**0.0006**	0.9717	**0.0362**	0.6108	**0.0009**

Bold (P<0.05), *Italic (P<0.10)*

SS=Subjective Score, ASF=Absolute Strain Force, MMD=Movement Measurement Device, ET=Exit Time

The CRH 240C>G genotype was a significant effect for MMD Peak 1, SS 3 and MMD Peak 3 (Table 4.16), while the interaction between CRH 240C>G and LEP was significant for ASF 1, ET 3 and ET 3-1 (Table 4.17), and the interaction between CRH 22C>G and LEP was significant for ASF 1 (Table 4.18). These interactions are presented graphically in Figures 4.8 through 4.11. For these figures, the values for each respective behaviour measurement have been separated into the average for each of the nine possible genotypes. The left three bars are the *LEP CC* animals, while the middle three bars are the *LEP CT* animals and the right three are the *LEP TT* animals. Within each group of three like coloured bars, the left-most bar is for the *CRH CC* animals, the middle bar is for the *CRH CG* animals and the right-most bar is for the *CRH GG* animals.

Table 4.16 LSM (±SEM) for behaviour measurements where there was a significant (P<0.05) effect of CRH 240C>G genotype

	CRH 240C>G		
	CC	CG	GG
MMD Peak 1 (P=0.0094)	21.817 (±6.3977) ^{ab}	27.902 (±4.6913) ^{b}	19.950 (±4.4150) ^{a}
SS 3 (P=0.0149)	2.097 (±0.0992) ^{ab}	2.217 (±0.0448) ^{b}	2.047 (±0.0377) ^{a}
MMD Peak 3 (P=0.0153)	80.888 (±9.5751) ^{ab}	94.080 (±5.8656) ^{b}	80.354 (±5.3708) ^{a}

^{ab} Means in the same row with different superscripts are significantly different (P<0.05)

SS=Subjective Score, MMD=Movement Measurement Device

Table 4.17 LSM (±SEM) for traits where the interaction between *LEP* & *CRH 240C>G* genotype was significant (P<0.05)

	ASF 1 (P=0.0158)			ET 3 (P=0.0005)			ET 3-1 (P=0.0004)		
	LEP genotype			*LEP* genotype			*LEP* genotype		
CRH 240C>G genotype	CC	CT	TT	CC	CT	TT	CC	CT	TT
CC	8643.05 (±22871)	77750 (±17824)	122447 (±28032)	15.567 (±2.4154)	4.352 (±2.0789)	2.864 (±2.9593)	12.146 (±2.3255)	0.401 (±1.8880)	-0.687 (±2.8006)
CG	85599 (±13110)	79241 (±10850)	87941 (±13735)	4.417 (±1.3998)	4.389 (±1.4482)	4.920 (±1.6584)	0.6947 (±1.2660)	0.621 (±1.2373)	0.976 (±1.4570)
GG	95413 (±11051)	85464 (±10186)	76497 (±13354)	4.086 (±1.2495)	4.6745 (±1.3221)	3.537 (±1.6222)	0.4209 (±1.1275)	0.622 (±1.1118)	-0.468 (±1.4072)

ASF=Absolute Strain Force, ET=Exit

Table 4.18 LSM (±SEM) for the trait where the interaction between *LEP* & *CRH 22C>G* genotype was significant (P<0.05)

	ASF 1 (P=0.0396)		
	LEP genotype		
CRH 22C>G genotype	CC	CT	TT
CC	83449 (±14450)	86121 (±12623)	75591 (±15614)
CG	69251 (±12930)	78342 (±11207)	91729 (±15455)
GG	36955 (±15759)	77992 (±12542)	119564 (±19639)

ASF=Absolute Strain Force

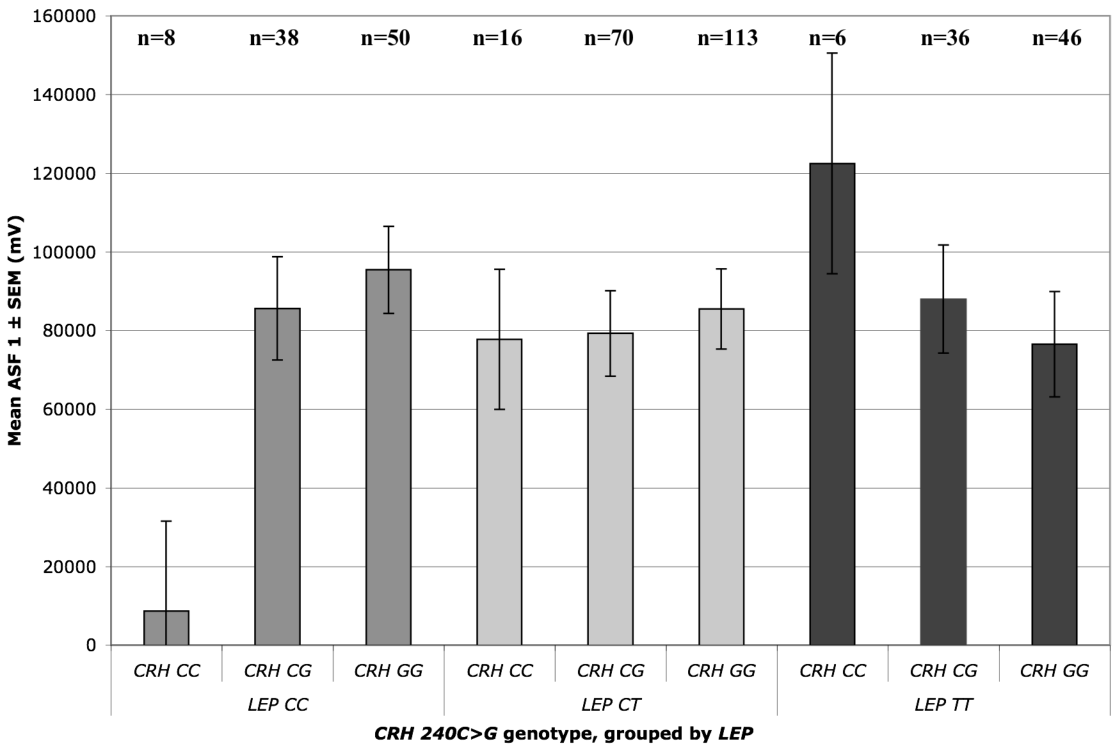


Figure 4.8 The interaction between *LEP* & *CRH 240C>G* genotypes for Absolute Strain Force 1

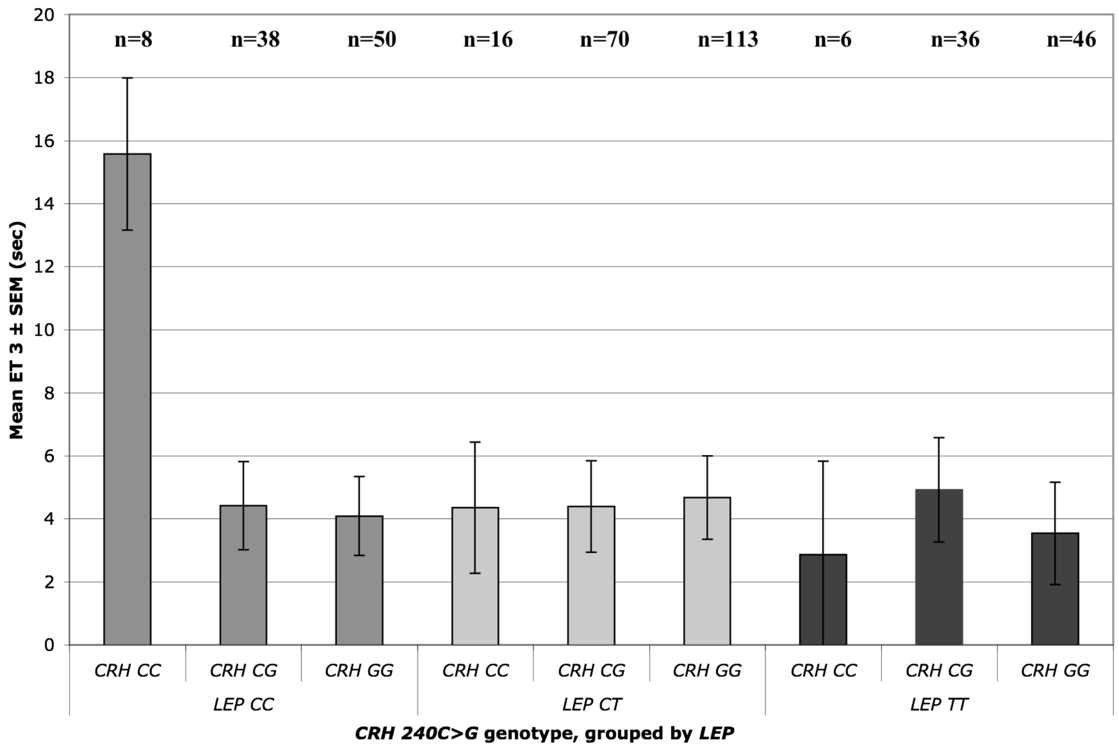


Figure 4.9 Interaction between *LEP* & *CRH 240C>G* genotypes for Exit Time 3

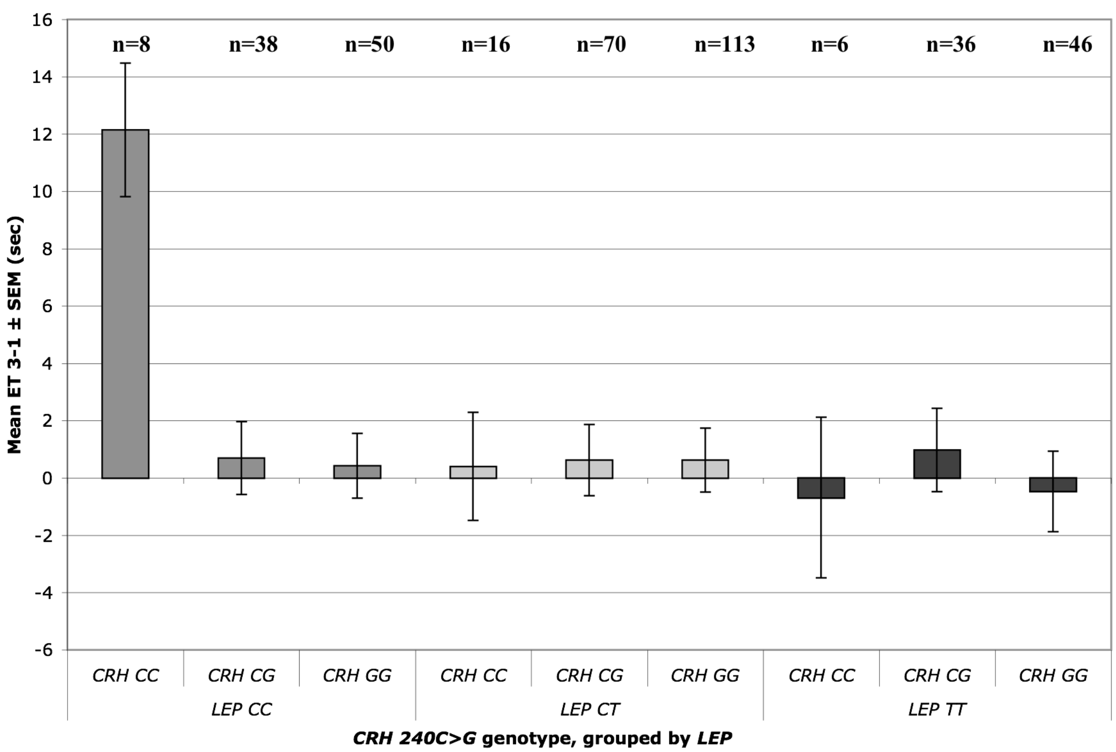


Figure 4.10 Interaction between *LEP* & *CRH 240C>G* genotypes for Exit Time 3-1

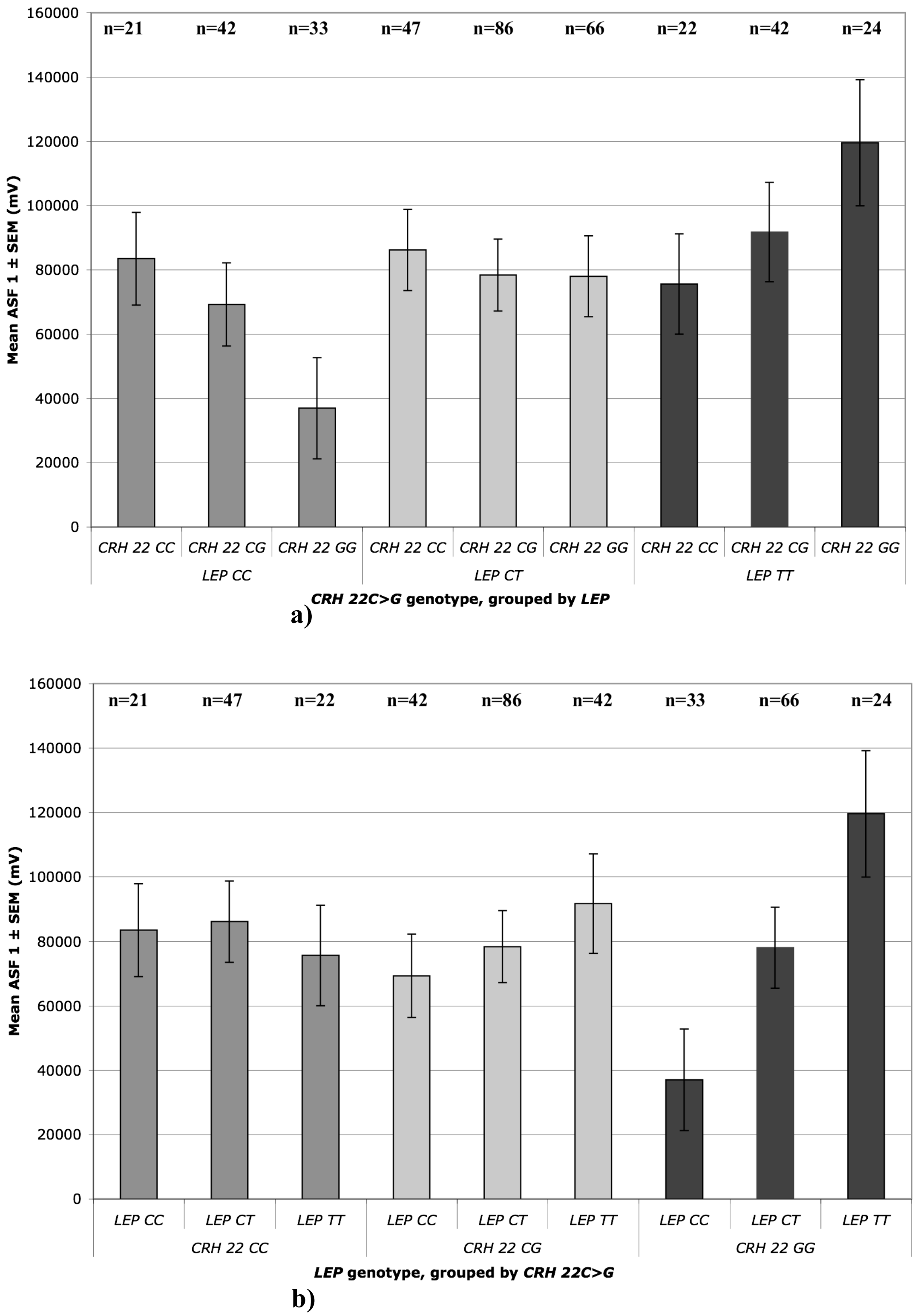


Figure 4.11 The interaction between *LEP* & *CRH 22C>G* genotypes for Absolute Strain Force 1. The upper graph (a) shows the data grouped by *LEP* genotype, while the lower graph (b) shows the same data grouped by *CRH 22C>G* genotype

4.3.3 Mixed model ANOVA: Carcass measurements & genotype

A summary of the results from the genotype and carcass measurements analysis is presented in Table 4.19.

Table 4.19 A summary of the tests of main effects for SNP genotypes on growth and carcass traits

Growth & Carcass traits	Test of Main Effects – P values				
	LEP	*CRH 22C>G*	*CRH 240C>G*	*LEP* x *CRH 22C>G*	*LEP* x *CRH 240C>G*
SOT Wt	0.7248	0.9381	0.4813	0.9402	0.6479
EOB Wt	0.9519	0.4422	*0.0712*	0.9909	0.8874
EOB ADG	0.9641	0.2023	0.1157	0.9989	0.9511
EOB BF	0.5520	0.6528	0.3327	0.2712	0.4924
EOB REA	0.4518	0.2017	0.3668	*0.1004*	0.8560
WC Wt	0.2798	0.5580	0.6652	0.7085	0.6534
Average fat	0.1472	0.3586	0.2161	0.7034	0.4792
Grade fat	0.3014	0.2205	*0.0784*	0.8905	0.5059
REA (cm^2)	0.6944	0.8251	0.2186	0.6699	0.1625
Marbling	0.8869	**0.0344**	0.1748	0.9123	0.6084
Cutability	0.2819	0.5082	0.5148	0.8815	0.1410
End of Finishing Wt	0.5600	0.6351	0.3818	0.9233	0.7347
Finishing ADG	0.2639	0.8557	0.9615	0.6770	0.8640

Bold (P<0.05), *Italic (P<0.10)*

SOT=Start of Test, WT=Weight, EOB=End of Backgrounding, ADG=Average Daily Gain, BF=Backfat,EA=Rib-eye Area, WC=Warm Carcass

5 DISCUSSION

5.1 Allele Frequencies

The allele frequencies found here (Table 4.1) are reasonably consistent with those previously reported for these SNPs. Buchanan *et al.* (2002a) reported overall *LEP* allele frequencies of C=0.54 and T=0.46 in a population of 154 beef bulls, with significant differences in the allele frequency between Angus (C=0.42) and Continental breeds (Charolais C=0.66 and Simmental C=0.68). Buchanan *et al.* (2005) reported allele frequencies for *CRH 22C>G* of C=0.37 and G=0.63, and for *CRH 240C>G* of C=0.29 and G=0.72 in Charolais cross steers. The allele frequencies reported here seem to indicate that there is a stronger influence of the British breeds. The *LEP* C allele frequency is lower in this population than the all breed total from Buchanan *et al.* (2002a). Additionally, the difference in *CRH 22C>G* allele frequencies indicate a different influence than the Charolais cross steers from Buchanan *et al.* (2005).

5.2 Behaviour Measurements & Habituation

The Shapiro-Wilk test for normality results, along with the skewness and Kurtosis values (Table 4.3) show that the behaviour measurement data did not have a normal distribution. We choose to use parametric statistical analysis despite the non-normal distribution mainly because of our large sample size. Willams and Nernez (1995) analyzed a sets of non-normally distributed test results using both parametric and non-parametric methods in order to determine if the different methods would result in different conclusions being drawn from the data. They found that in their large sample size subset (n=356), the parametric and non-parametric methods lead to the same conclusions, although the P values generated were slightly different. We feel that these results justify our use of parametric analyses for our data.

We found no indications that the steer's weight had an effect on any of the behaviour measurements. For ASF, we used an estimated weight on the day of measurement as a covariate, as we felt that there was a possibility of weight having an effect on the strain gauge values. For the other behaviour measurements, we used start of test weight as a covariate. None of the weight variables were a significant effect, therefore they were left out of all subsequent analyses.

The majority of the steers examined in this study had fairly calm temperaments, which is the most obvious in the SS (Table 4.2). The average SS was around 2 on the 1-5 scale found here is in good agreement with previous studies of a similar nature. For example, Grandin (1993) found that 64% of steers they studied had a SS of 2 or less over 3 measurement sessions, and Voisinet *et al.* (1997) found a mean SS of 1.8 in *B. taurus* steers using the same scale. Sebastian *et al.* (2007) performed detailed analyses on the relationships between the various behaviour measurements used in this study, and found consistent correlations between the SS and other measurements. This indicates that although SS is a fundamentally less desirable and less reliable measure than the objective measurements, it is none the less valuable for giving an overall impression of the steers' temperament.

Habituation, *i.e.* an improvement in a particular behaviour over time, was observed only for ASF and ET and not for SS or MMD Peaks (Table 4.4). This is in contrast to Schmutz *et al.* (2001), who found habituation in MMD scores in their QTL study of temperament. It is likely that the difference between these results can be explained by the number of times the cattle were exposed to the handling facilities in each trial. In Schmutz *et al.* (2001) the cattle were weighed every other week for several months, while here they were only through the facility 6 times and at ~1 month intervals during the behaviour measurement period (Table 3.1). One other difference that has the potential to affect the results is, that in the Schmutz *et al.* (2001) study, the calves were temperament tested for the first time immediately after being unloaded at the Beef Research Facility, while in this study the first behaviour measurement was the day after the cattle had been purchased, transported and processed. It is therefore possible that our measurement values in Session 1 were simply uncharacteristically low due to an exhaustion factor from the rigours and strain of the previous day. It is also possible the initial temperament scores in the Schmutz *et al.* (2001) study were higher since that measurement was far more closely related to the agitation of the weaning processes.

The delay between arrival and the first measurement session in this current study was deliberate, and intended to ensure that all of the steers' most recent experiences were as similar as possible. Because the steers used here were from an unknown number of farms, it is possible that their different experiences prior to arrival at the U of S facility could have had an impact on their behavioural responses. In Schmutz *et al.* (2001) the animals originated from the same breeding program, and were kept on pasture at one research station for several months.

It is interesting to consider that while Burrow (1997) noted that most tests he reviewed indicated a general improvement in cattle temperament with age and experience, Grandin (1993) concluded that cattle temperament is relatively stable over a 5 month period and recommended that breeding or culling decisions based on temperament should consider at least 3 measurement sessions. This may indicate that habituation to handling requires a longer term and considerably more repetition than was afforded by the particular experimental design employed in this trial.

At least two recent studies have specifically examined the change in ET or "flight speed" over a series of measurements. Müller *et al.* (2006) showed that flight speed increased, which is less desirable, over 3 measurements at four week intervals in Angus heifers. In a study with similar results to those presented here, Curley *et al.* (2006) showed that average exit velocity for Brahman bulls decreased over 3 measurements taken at 30 day intervals. It is unclear why ET habituation appears to be somewhat inconsistent between studies with similar set ups and measurement intervals. It is possible that the early experiences of the subject animals had a large influence on habituation, therefore it may be difficult to get consistent results between studies with animals reared under different conditions. It is also possible that other variables such as time of day, weather or season may have an influence on ET measurements, and therefore makes results difficult to reproduce. For example, in this study there was one measurement day where we used grit on a patch of ice in the ET chute, and it did appear to cause some of the cattle to pause slightly.

5.3 Associations of Genotype with Growth and Carcass Measurements

The number of T alleles at the *LEP* SNP showed positive correlations with EOT BF, Avg. Fat and Grade fat (Table 4.10), which is in good agreement with previous reports of associations between the *LEP* T allele and fatter carcasses (Buchanan *et al.* 2002a, Kononoff *et al*. 2005, Buchanan *et al.* 2007). The negative correlation between the number of T alleles and cutability (Table 4.10) also fits the established pattern, as carcasses with higher fat content have decreased lean yield.

Regression analysis for the *CRH 22C>G* G allele (Table 4.11) did not corroborate the previously reported correlations with increased REA or HCW (Buchanan *et al.* 2005).

For the *CRH 240C>G* SNP, the carcass results (Table 4.12) are similar to previous findings. Buchanan *et al.* (2002b) showed an association between the C allele and increased weaning weight and yearling weight EPDs. In this study, we found that the G allele showed a negative correlation with REA and a trend in the same direction for ultrasound REA, which would equate to the C allele being associated with increased growth. Buchanan *et al.* (2005) also showed that the *CRH 240C>G* SNP was associated with WCW.

When the genotypes were analyzed as main effects on the growth and carcass traits using mixed model ANOVA, few significant results were found (Table 4.19), and these results did not support the previous growth and carcass associations (Buchanan *et al.* 2002a, Kononoff *et al*. 2005, Buchanan *et al.* 2005, Buchanan *et al.* 2007). It is unclear at this point why there were no significant associations between genotype and carcass traits in this population.

5.4 Behaviour & Genotype

The simple regression results indicated that an increased number of *LEP T* alleles was associated with calmer temperament as measured by MMD Peaks (Table 4.7). An increased number of *CRH 240C>G G* alleles was also associated with calmer temperament as measured by MMD Peaks in the first session (Table 4.9), and the *CRH 240C>G G* allele was favorable for ET improvement over time. These simple regression results, while interesting as preliminary analyses, are less meaningful than the ANOVA analyses that followed.

The SNP genotypes were significant main effects on several of the behaviour measurements in the ANOVA analyses. Table 4.14 shows the significant main effects of the *CRH 240C>G* SNP. For MMD Peak 1, SS 3 and MMD Peak 3, *GG* animals were significantly different from *CG* but not *CC* animals, and *CC* and *CG* were not significantly different. These results are difficult to explain, and do not seem to follow any pattern that would indicate an advantage of one SNP allele over the other.

The remaining significant effects of genotypes on behaviour measurements consist of interactions between the *LEP* and each of the *CRH* SNPs. For ASF 1 (Figure 4.8), ET 3 (Figure 4.9) and ET 3-1 (Figure 4.10), CC at both *LEP* and *CRH 240C>G* appears to be associated with the cattle with the calmest temperaments. In the ASF measurement, a lower value indicates an animals that struggles less in the squeeze chute, and thus the lower value of the CC/CC *LEP/CRH 240C>G* genotype is favorable. For the ET measurement, a higher value indicates a calmer animal that was moving more slowly out of the exit chute, and therefore the CC/CC *LEP/CRH 240C>G* genotype is once again favorable. For the ET 3-1 habituation calculation, a higher value indicates a larger improvement in ET between sessions 1 and 3, and the cattle of the CC/CC *LEP/CRH 240C>G* genotype showed the most improvement in ET.

The single significantly different genotype seems to indicate that the favorable results could be a double recessive trait (CC/CC *LEP/CRH 240C>G*). However, visual examination of Figures 4.8, 4.9 and 4.10 could also suggest that there may instead be an additive effect of *CRH 240C>G* genotype, with opposite effects in *LEP CC* and *LEP TT* animals.

One possible confounding factor in the interactions between *LEP* and *CRH 240C>G* is that the numbers of animals of each genotype are uneven – there are only eight with the CC/CC *LEP/CRH 240C>G* genotype, and only six *TT/CC*. Should this research be repeated, it may be valuable to genotype the cattle prior to weaning and the behaviour measurement process, and select animals for the trial so that there is a more equal number of steers of each genotype.

The final significant interaction is between *LEP* and *CRH 22C>G* for ASF 1 (Figure 4.11). The ASF 1 values are only significantly different between *LEP* genotypes in *CRH 22C>G GG* animals, where there appears to be an additive effect of the *LEP C* allele on the ASF 1 values.

The *LEP CC* genotype appears to be associated with the calmest animals as measured by ASF 1 and ET 3 (Table 4.13). This is slightly disconcerting, as some cattle producers are selecting breeding stock based on the previous associations of *LEP TT* with increased fat and better quality and yield grades (Buchanan *et al*. 2002a; Kononoff *et al*. 2005; Buchanan *et al*. 2007), and it is possible that selecting for *LEP TT* could inadvertently select for poorer temperaments.

However, the interactions presented here indicate that *LEP* genotype should be considered in conjunction with *CRH 22C>G* and *CRH 240C>G* genotypes, and that the *LEP TT* can really be considered to be of "normal" temperament. This conclusion is based on the fact that the *LEP CC* genotype was significantly better than the others, but that there were no differences between *LEP CT* and *TT*.

While it is acknowledged that the results presented here should be validated in additional populations, they do indicate that producers may be able to use these SNPs to aid in selection for temperament traits. For example, should a producer choose to attempt selection for the calmest ASF 1, ET 3 or ET 3-1 traits, they could select animals that are *LEP* CC and *CRH 240C>G CC*, or *LEP* CC and *CRH 22C>G GG* for ASF 1. However, should they choose to select for the *LEP TT* genotype, the potential for a negative effect on ASF 1 could be avoided by selecting for animals that are any *CRH 22C>G* genotype other than *GG*. The LEP and *CRH 240C>G* interaction for ASF 1, ET 3 and ET 3-1 indicates that although *CC/CC* seems to be favorable, there are no significant differences between the other genotype combinations, therefore selecting *LEP TT* animals is not likely to negatively influence these traits.

Before we begin to recommend selection for temperament based on the associations with the SNPs discussed here, it would be prudent to understand physiological effects of the SNPs and their interactions. We do know that both *LEP* and *CRH* are involved in the HPA axis, and that all three SNPs studied result in amino acid changes in the respective hormones. It has been previously shown that *LEP TT* cattle express higher levels of leptin mRNA than *LEP CC* cattle (Buchanan *et al*. 2002a). The effects of the *CRH* SNPs on *CRH* expression have not been examined, neither has whether the *LEP* or *CRH* SNPs affect the gene's functions in the HPA axis. Unfortunately, such an examination was beyond the scope of this project.

6 CONCLUSIONS

The purpose of the study reported here was to determine if SNPs in *LEP* and *CRH* can be associated with temperament measurements in cattle. We have shown that there are significant interactions between *LEP* and *CRH* genotypes for several objective measurements of cattle response to handling. Producers may be able to use these associations to select for favorable temperaments, and thereby potentially reduce the frequency of injuries to their cattle and personnel. For all of the interactions reported with the behaviour measurements, the *LEP CC* genotype appears to associated with the calmest responses. However, if *LEP* genotype is considered along with the *CRH* genotypes according to the statistical interaction reported, and we consider *LEP* CT and TT animals to have normal temperaments, it should be possible to select for the T allele for fatness without detrimental behavioral effects.

The results of this project suggest considerable potential for further work. One project that is beginning is the use of RIA to determine if there are differences in plasma cortisol between animals of different *LEP* and *CRH* genotypes, and whether those differences are reflected in the various behaviour measurements. The existing DNA samples and behaviour data could also be used to search for associations with mutations in other genes involved in the HPA axis, such as the CRH receptors, ACTH and arginine vasopressin, among many others.

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APPENDICES

Appendix A   Backgrounding Diet

Ingredient (%, as fed)	Dates fed		
	11/29/05-12/13/05	12/14/05-03/20/06	03/21/06-05/02/06
Barley Silage	40	50	50
Barley Grain	20	25	25
Pellet	5*	5**	5**
Hay	35	10	20
Straw	0	10	0

*JM Starter   **RB1

Ingredient (%, as fed)	Pellet Name	
	JM Starter	RB1
Barley Grain	7	-
Tallow	3	-
Molasses	3.5	3.2
Canola Meal	61	68.3
Ground Limestone	6.5	10
TM Salt	4.5	4.5
JM Rumensin Premix*	5.5	5
Lab Supplement 106**	9	9

*Contains 97% barley grain and 3% Rumensin

** Contains barley grain mixed with Vitamins A & D

Appendix B   Finishing Diet

Ingredient	% as fed
Barley Grain	66.8
Barley Silage	17
Wet Distillers Grain*	10
Supplement	4.2
Grass Hay	2

*the steers also had access to thin stillage as a fluid source

Appendix C Steer Genotypes at LEP, CRH 22C>G & CRH 240C>G SNPs

Lab #	Tag #	*LEP*	*CRH 22C>G*	*CRH 240C>G*
06-001	12	TT	CG	GG
06-002	7	CT	CG	CG
06-003	5	CC	CG	CG
06-004	20	CC	GG	GG
06-005	25	CC	GG	GG
06-006	19	CC	GG	GG
06-007	23	CT	CG	CG
06-008	18	CC	CG	CG
06-009	27	CT	CG	CG
06-010	3	CT	CG	CG
06-011	21	TT	GG	GG
06-012	33	CT	CG	GG
06-013	26	CC	CC	CC
06-014	4	CT	CC	CG
06-015	15	CC	CG	CG
06-016	17	CT	GG	GG
06-017	14	CT	CG	CG
06-018	29	CT	CC	CC
06-019	13	CC	CG	CG
06-020	2	TT	CG	GG
06-021	16	CT	GG	GG
06-022	8	CT	CG	CG
06-023	1	CT	GG	GG
06-024	10	CT	GG	GG
06-025	30	CT	CG	GG
06-026	28	CC	CC	CC
06-027	32	CT	CG	CG
06-028	9	CT	GG	GG
06-029	31	CC	CG	GG
06-030	24	TT	CC	CG
06-031	22	CC	CG	CG
06-032	11	CC	GG	GG
06-033	6	CT	GG	GG
06-034	65	CC	GG	GG
06-035	41	TT	GG	GG
06-036	39	CC	CG	CG
06-037	64	CT	CG	CG
06-040	50	TT	GG	GG
06-041	55	CC	CG	CG
06-042	52	TT	CC	GG
06-043	54	CC	CG	GG
06-044	51	CC	GG	GG
06-045	48	CT	CG	GG
06-046	40	CC	GG	GG
06-047	45	CT	CG	GG
06-048	43	CT	CG	CG
06-049	49	CC	CG	GG
06-050	61	CT	CG	GG
06-051	59	CT	GG	GG
06-052	38	CC	CG	CG
06-053	46	CC	CG	GG
06-054	53	CT	CG	CG
06-055	67	CT	GG	GG
06-056	56	CT	GG	GG
06-057	47	CC	CG	CG
06-058	57	TT	CC	CG
06-059	63	CT	GG	GG
06-060	58	CT	GG	GG
06-061	34	CC	CC	CG
06-062	42	CT	GG	GG
06-063	62	CC	CG	GG
06-064	35	CT	CG	CG
06-065	36	CC	GG	GG
06-066	60	CT	GG	GG
06-067	44	CT	CG	GG
06-068	66	CC	GG	GG
06-069	37	CT	GG	GG
06-070	76	CT	CC	GG
06-071	81	TT	CG	CG
06-072	78	CC	CC	GG
06-073	97	CC	CC	CC
06-074	79	CT	CG	GG
06-075	86	TT	CG	GG
06-076	96	CT	CC	CG
06-077	68	CT	CG	GG
06-078	87	CC	CG	CG
06-079	93	CT	CG	CG
06-080	69	CC	GG	GG
06-081	98	CT	CC	CC
06-082	71	CC	CG	CG
06-083	99	CC	CG	GG
06-084	94	CC	CC	CG

Lab #	Tag #	*LEP*	*CRH* *22C>G*	*CRH* *240C>G*
06-085	90	CC	CG	GG
06-086	95	TT	CG	CG
06-087	73	TT	CG	GG
06-088	70	CC	GG	GG
06-089	77	CC	GG	GG
06-090	85	CC	CG	CG
06-091	83	CT	CC	GG
06-092	82	CC	CG	GG
06-093	84	CC	CG	CG
06-094	91	CT	GG	GG
06-095	88	CT	CG	CG
06-096	72	CT	CG	CG
06-097	89	TT	CC	CC
06-098	100	CT	GG	GG
06-099	92	TT	CG	CG
06-100	80	TT	CG	CG
06-101	75	CT	CG	GG
06-102	110	CT	GG	GG
06-103	109	CC	GG	GG
06-104	116	CT	CG	CG
06-105	118	CT	CG	CG
06-106	133	CT	CC	GG
06-107	126	CC	CC	CC
06-108	112	CT	GG	GG
06-109	120	TT	GG	GG
06-110	124	TT	CC	CG
06-111	104	CT	GG	GG
06-112	113	CC	CC	CC
06-113	122	CT	CG	CG
06-114	121	TT	CG	CG
06-115	117	CC	CG	CG
06-116	115	CT	GG	GG
06-117	129	CT	GG	GG
06-118	130	CC	GG	GG
06-119	127	CC	GG	GG
06-120	132	CT	CG	GG
06-121	108	CT	CG	GG
06-122	106	CC	GG	GG
06-123	105	TT	CG	CG
06-124	101	CC	CG	CG
06-125	119	TT	GG	GG
06-126	111	CT	CG	CG
06-127	123	CC	GG	GG
06-128	125	CT	GG	GG
06-129	128	TT	GG	GG

Lab #	Tag #	*LEP*	*CRH* *22C>G*	*CRH* *240C>G*
06-130	107	CT	CG	CG
06-131	102	CT	GG	GG
06-132	131	CC	CG	CG
06-133	114	CT	CC	CG
06-134	103	CT	GG	CC
06-135	74	CT	CG	GG
06-136	147	TT	CC	GG
06-137	164	CC	GG	GG
06-138	146	CT	CC	GG
06-139	162	TT	CC	CC
06-140	144	CT	CC	CC
06-141	163	CC	CG	CG
06-142	148	CT	CG	GG
06-143	158	CT	CG	CG
06-144	140	TT	CG	CG
06-145	145	TT	GG	GG
06-146	136	CT	CG	GG
06-147	155	CT	CG	GG
06-148	142	CC	CG	CG
06-149	152	CT	CC	CG
06-150	159	CC	CG	CG
06-151	138	TT	CG	CG
06-152	139	CT	GG	GG
06-153	156	CC	GG	GG
06-154	135	TT	GG	GG
06-155	137	CC	GG	GG
06-156	166	CT	GG	GG
06-157	165	CT	CG	CG
06-158	149	CT	CG	CG
06-159	151	TT	CG	CG
06-160	143	CT	CC	CC
06-161	161	CT	GG	CG
06-162	160	CC	GG	GG
06-163	141	CT	GG	CG
06-164	153	CC	CG	CG
06-165	134	CT	CC	CC
06-166	154	CT	CG	CG
06-167	157	CT	CG	CG
06-168	150	CC	CG	CG
06-169	221	CC	CG	CG
06-170	218	TT	GG	GG
06-171	202	TT	GG	GG
06-172	201	TT	CG	CG
06-173	223	CT	CC	CC
06-174	203	CT	GG	GG

Lab #	Tag #	*LEP*	*CRH 22C>G*	*CRH 240C>G*
06-175	216	CT	GG	GG
06-176	211	CC	CG	GG
06-177	210	CC	CC	GG
06-178	215	CT	CG	CG
06-179	200	CT	GG	GG
06-180	209	CC	GG	GG
06-181	212	CT	CG	GG
06-182	206	CT	CC	GG
06-183	225	CT	CG	GG
06-184	229	TT	CG	CG
06-185	204	CT	CG	CG
06-186	222	CT	GG	GG
06-187	219	TT	CG	GG
06-188	205	CC	GG	GG
06-189	213	CT	CG	CG
06-190	208	CC	GG	GG
06-191	224	TT	CC	CC
06-192	226	CT	GG	GG
06-193	217	TT	CG	CG
06-194	227	CT	CC	CG
06-195	207	TT	GG	GG
06-196	220	CT	CG	GG
06-197	214	CT	CG	GG
06-198	228	CC	GG	GG
06-199	180	TT	CG	GG
06-200	184	CC	GG	GG
06-201	188	CT	CG	GG
06-202	186	CT	GG	GG
06-203	194	CT	CG	CG
06-204	179	TT	CG	CG
06-205	174	CC	CG	GG
06-206	170	CT	CC	CG
06-207	185	CC	CC	GG
06-208	196	CC	CC	GG
06-209	182	TT	CG	GG
06-210	171	CT	CG	GG
06-211	183	CT	CC	CG
06-212	173	TT	CG	CG
06-213	181	CT	CG	GG
06-214	191	CT	CG	CG
06-215	187	CT	GG	GG
06-216	199	TT	CC	CG
06-217	197	CC	CG	CG
06-218	193	CC	CC	CG
06-219	189	CT	GG	GG
06-220	178	CT	GG	GG
06-221	168	TT	CG	GG
06-223	195	TT	CC	CG
06-224	167	CT	CC	CC
06-225	175	CT	CG	GG
06-226	169	CT	GG	GG
06-227	177	CT	GG	GG
06-228	192	CT	GG	GG
06-229	176	CT	GG	CG
06-230	198	CT	CG	GG
06-231	190	CT	GG	GG
06-232	172	TT	CC	GG
06-233	237	TT	GG	GG
06-234	258	TT	CC	CG
06-235	265	CC	GG	GG
06-236	249	CT	CG	GG
06-237	236	CC	CG	GG
06-238	256	CT	GG	GG
06-239	238	CT	CC	CG
06-240	239	TT	CG	CG
06-241	230	TT	CG	CG
06-242	232	CT	GG	GG
06-243	233	CT	CC	CG
06-244	251	CT	CG	GG
06-245	259	TT	GG	GG
06-246	243	CT	CG	GG
06-247	231	CT	CC	CC
06-248	252	TT	GG	GG
06-249	262	CT	GG	GG
06-250	253	CT	GG	GG
06-251	246	TT	CG	CG
06-252	241	CT	GG	GG
06-253	254	CT	GG	GG
06-254	257	CC	GG	CG
06-255	267	TT	CG	CG
06-256	242	CT	CG	GG
06-257	247	CC	CG	CG
06-258	255	CT	CG	CG
06-259	260	CC	CG	CC
06-260	234	CC	CC	CG
06-261	264	CC	CG	CG
06-262	266	CC	GG	GG
06-263	235	CT	GG	GG
06-264	268	CT	CG	CG
06-265	250	TT	CG	GG

Lab #	Tag #	*LEP*	*CRH 22C>G*	*CRH 240C>G*
06-266	334	TT	CG	CG
06-267	330	CT	CG	CG
06-268	314	TT	GG	GG
06-269	333	CT	CG	CG
06-270	328	CT	CC	CG
06-271	322	CC	CG	CG
06-272	307	TT	CG	CG
06-273	336	CC	CC	CC
06-274	337	CC	GG	GG
06-275	339	TT	CG	GG
06-276	315	TT	CC	CC
06-277	326	CT	CC	CC
06-278	309	CT	CG	GG
06-279	325	CT	CG	CG
06-280	327	CC	CG	CG
06-281	317	CT	GG	GG
06-282	320	CT	GG	GG
06-283	319	TT	CC	CG
06-284	323	TT	CC	CG
06-285	335	CC	CC	CC
06-286	312	TT	GG	GG
06-287	340	TT	CC	CG
06-288	324	TT	GG	GG
06-289	329	CT	GG	GG
06-290	308	CT	CC	CG
06-291	311	CT	CC	CG
06-292	313	CT	CG	CG
06-293	310	TT	CC	CG
06-294	332	CT	CC	CG
06-295	331	TT	CG	CG
06-296	316	TT	CG	GG
06-297	318	TT	CG	CG
06-298	338	TT	GG	GG
06-299	321	CT	GG	GG
06-300	296	CT	CG	GG
06-301	301	CT	CG	GG
06-302	294	CT	CC	GG
06-303	292	CT	GG	GG
06-304	289	CT	CG	CG
06-305	263	CT	CG	GG
06-306	290	TT	GG	GG
06-307	277	CT	GG	GG
06-308	304	CT	CG	CG
06-309	281	TT	GG	GG
06-310	293	CT	GG	GG

Lab #	Tag #	*LEP*	*CRH 22C>G*	*CRH 240C>G*
06-311	269	CT	CG	GG
06-312	240	TT	CG	CG
06-313	288	CT	GG	GG
06-314	274	TT	GG	GG
06-315	272	TT	CG	GG
06-316	271	TT	CG	GG
06-317	306	CT	CG	GG
06-318	248	CT	CG	CG
06-319	287	CT	CC	CC
06-320	280	CT	CC	CG
06-321	285	CC	CC	CG
06-322	283	CC	CG	CG
06-323	295	CC	CG	GG
06-324	245	CT	CC	CG
06-325	278	CT	CC	CG
06-326	291	CC	CC	GG
06-327	300	CC	CC	GG
06-328	270	CC	CC	GG
06-329	305	CT	CC	GG
06-330	303	CT	CG	GG
06-331	273	CT	CC	CG
06-332	261	CT	GG	GG
06-333	275	CT	CG	CG
06-334	348	CT	CG	CG
06-335	366	CT	CG	GG
06-336	365	CC	CG	CG
06-337	350	TT	CG	GG
06-338	345	TT	CG	GG
06-339	359	CT	GG	GG
06-340	347	TT	CC	CC
06-341	361	CT	CG	GG
06-342	346	CT	CG	GG
06-343	372	CT	CG	GG
06-344	356	CT	CG	CG
06-345	362	CT	CG	CC
06-346	369	CT	CG	CG
06-347	352	CT	CC	CG
06-348	374	CT	CC	CC
06-349	373	CT	CC	CG
06-350	355	CT	CG	GG
06-351	349	CT	CG	CG
06-352	353	TT	CG	GG
06-353	343	TT	CG	GG
06-354	360	CT	CC	CG
06-355	354	CT	CG	GG

Lab #	Tag #	*LEP*	*CRH* 22C>G	*CRH* 240C>G
06-356	351	TT	GG	GG
06-357	367	CT	GG	GG
06-358	370	CT	CC	CG
06-359	341	CT	CG	CG
06-360	358	TT	CC	CG
06-361	371	TT	CG	CG
06-362	344	CT	GG	GG
06-363	342	CT	CC	CC
06-364	363	CT	CC	CC
06-365	364	TT	CC	CC
06-366	357	CT	CC	CC
06-367	298	CC	GG	CG
06-368	279	CT	CG	CG
06-369	385	CT	CC	CG
06-370	276	TT	CG	GG
06-371	384	CT	CC	GG
06-372	388	CT	GG	GG
06-373	394	CT	GG	GG
06-374	284	CC	GG	GG
06-375	380	CT	GG	GG
06-376	397	TT	CC	CG
06-377	282	CT	CG	GG
06-378	378	TT	GG	GG
06-379	375	CC	GG	GG
06-380	400	CT	GG	GG
06-381	379	CC	CG	CG
06-382	386	CT	GG	GG
06-383	286	CT	GG	GG
06-384	391	CT	GG	GG
06-385	377	CT	GG	GG
06-386	398	TT	GG	GG
06-387	390	TT	CG	GG
06-388	299	CT	CC	CC
06-389	399	CT	CG	CG
06-390	382	CC	CC	CG
06-391	376	CC	CC	CG
06-392	383	CT	CC	GG
06-393	297	CT	CC	CG
06-394	393	CT	GG	GG
06-395	381	TT	CG	CG
06-396	389	CT	CG	GG
06-397	396	CT	CC	CG
06-398	387	TT	GG	GG
06-399	302	CT	CC	CG
06-400	395	TT	CC	CG

Lab #	Tag #	*LEP*	*CRH* 22C>G	*CRH* 240C>G
06-401	392	CT	CG	CG

Appendix D Behaviour Measurements

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-001	12	2	152308.40	14	2.83	1	47660.00	105	1.35	-1.00	-104648.40	91.00	-1.48
06-002	7	2	105510.20	34	3.03	2	114707.00	13	2.90	0.00	9196.80	-21.00	-0.13
06-003	5	2	52006.15	0	4.72	2	32471.00	5	4.00	0.00	-19535.15	5.00	-0.72
06-004	20	2	72461.47	84	2.68	2	71812.00	67	2.69	0.00	-649.47	-17.00	0.01
06-005	25	1	76849.86	19	4.22	2	66542.00	24	2.42	1.00	-10307.86	5.00	-1.80
06-006	19	2	46108.99	11	4.81	2	57945.00	21	17.26	0.00	11836.01	10.00	12.45
06-007	23	3	.	52	3.31	2	45079.00	198	1.48	-1.00	.	146.00	-1.83
06-008	18	1	36592.48	63	7.50	2	85850.00	4	43.25	1.00	49257.53	-59.00	35.75
06-009	27	1	27544.93	19	4.70	1	39081.00	39	16.78	0.00	11536.07	20.00	12.08
06-010	3	1	52986.19	17	3.54	2	102083.00	81	3.86	1.00	49096.81	64.00	0.32
06-011	21	2	69511.71	2	4.75	2	112197.00	67	2.94	0.00	42685.29	65.00	-1.81
06-012	33	1	57940.80	13	4.24	2	120459.00	75	3.55	1.00	62518.20	62.00	-0.69
06-013	26	1	3928.96	28	2.71	2	23773.00	115	3.17	1.00	19844.04	87.00	0.46
06-014	4	2	162486.40	2	3.53	3	124137.00	23	2.77	1.00	-38349.40	21.00	-0.76
06-015	15	2	113844.50	98	1.95	3	71532.00	204	1.33	1.00	-42312.50	106.00	-0.62
06-016	17	1	4998.65	9	14.49	2	26840.00	65	2.87	1.00	21841.35	56.00	-11.62
06-017	14	2	85380.58	42	2.37	2	.	138	3.69	0.00	.	96.00	1.32
06-018	29	1	66597.36	9	3.14	2	66162.00	95	7.22	1.00	-435.36	86.00	4.08
06-019	13	2	102969.20	47	2.32	1	19420.00	78	2.73	-1.00	-83549.20	31.00	0.41
06-020	2	1	.	1	3.08	2	141480.00	20	3.49	1.00	.	19.00	0.41
06-021	16	1	27030.41	37	3.68	2	99258.00	82	2.48	1.00	72227.59	45.00	-1.20
06-022	8	2	106053.77	22	3.69	2	121919.00	70	2.48	0.00	15865.23	48.00	-1.21
06-023	1	1	57977.04	0	3.07	2	73190.00	40	8.03	1.00	15212.96	40.00	4.96
06-024	10	2	99222.29	30	3.99	2	16086.00	57	3.49	0.00	-83136.29	27.00	-0.50
06-025	30	2	29181.62	59	3.26	3	88181.00	88	4.82	1.00	58999.38	29.00	1.56
06-026	28	2	76083.10	17	3.99	2	214441.00	105	6.45	0.00	138357.90	88.00	2.46
06-027	32	2	234191.40	11	3.83	2	86895.00	40	3.51	0.00	-147296.40	29.00	-0.32
06-028	9	2	77420.47	13	3.63	2	139625.00	11	3.17	0.00	62204.53	-2.00	-0.46
06-029	31	2	21827.73	14	2.79	2	92140.00	98	2.38	0.00	70312.27	84.00	-0.41

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-030	24	1	27957.05	2	3.60	2	70010.00	63	3.20	1.00	42052.96	61.00	-0.40
06-031	22	2	97322.90	39	1.29	2	128323.00	181	1.28	0.00	31000.10	142.00	-0.01
06-032	11	2	117141.50	48	5.01	2	120575.00	122	7.07	0.00	3433.50	74.00	2.06
06-033	6	2	125243.80	14	3.93	2	122386.00	104	1.67	0.00	-2857.80	90.00	-2.26
06-034	65	2	95356.42	34	4.09	2	173020.00	137	3.25	0.00	77663.58	103.00	-0.84
06-035	41	1	41309.10	0	4.62	2	169454.00	76	6.73	1.00	128144.90	76.00	2.11
06-036	39	2	119908.20	24	2.79	1	36572.00	76	2.23	-1.00	-83336.20	52.00	-0.56
06-037	64	1	23852.70	20	3.32	2	77072.00	83	4.04	1.00	53219.30	63.00	0.72
06-040	50	3	153216.87	68	2.71	3	163477.00	125	4.04	0.00	10260.13	57.00	1.33
06-041	55	2	87273.40	0	4.53	3	110281.00	15	3.57	1.00	23007.60	15.00	-0.96
06-042	52	1	22431.82	4	3.28	2	25834.00	108	1.99	1.00	3402.18	104.00	-1.29
06-043	54	2	53202.80	5	5.28	2	114013.00	9	8.89	0.00	60810.20	4.00	3.61
06-044	51	1	26322.02	3	3.15	2	67484.00	30	2.42	1.00	41161.98	27.00	-0.73
06-045	48	2	41195.99	15	3.21	1	45308.00	70	2.23	-1.00	4112.01	55.00	-0.98
06-046	40	1	46141.91	14	4.30	1	38751.00	104	4.14	0.00	-7390.91	90.00	-0.16
06-047	45	1	45972.60	16	5.47	2	158731.00	47	10.04	1.00	112758.40	31.00	4.57
06-048	43	2	59340.69	44	1.91	2	114757.00	168	2.13	0.00	55416.31	124.00	0.22
06-049	49	1	83774.50	1	2.87	2	73277.00	29	5.73	1.00	-10497.50	28.00	2.86
06-050	61	2	64627.12	0	3.60	2	163416.00	122	4.18	0.00	98788.88	122.00	0.58
06-051	59	2	217475.50	33	2.89	3	348595.00	135	1.36	1.00	131119.50	102.00	-1.53
06-052	38	3	65659.07	133	1.05	4	62333.00	286	1.37	1.00	-3326.07	153.00	0.32
06-053	46	3	173392.40	80	1.43	3	176077.00	143	1.36	0.00	2684.60	63.00	-0.07
06-054	53	2	19538.53	0	5.32	1	82880.00	19	5.82	-1.00	63341.47	19.00	0.50
06-055	67	2	97145.91	22	2.56	2	113833.00	119	2.65	0.00	16687.09	97.00	0.09
06-056	56	2	57859.02	26	3.25	2	54211.00	151	2.34	0.00	-3648.02	125.00	-0.91
06-057	47	2	77448.24	11	2.80	2	89350.00	155	2.11	0.00	11901.76	144.00	-0.69
06-058	57	2	153874.93	15	3.90	1	49907.00	39	2.69	-1.00	-103967.93	24.00	-1.21
06-059	63	2	67272.30	25	4.07	1	29455.00	68	3.75	-1.00	-37817.30	43.00	-0.32
06-060	58	1	111955.00	15	4.48	2	47385.00	57	9.27	1.00	-64570.00	42.00	4.79
06-061	34	2	153824.39	52	4.67	2	98496.00	95	2.39	0.00	-55328.39	43.00	-2.28
06-062	42	2	37042.80	1	3.74	2	92423.00	53	2.07	0.00	55380.20	52.00	-1.67
06-063	62	2	22324.87	27	3.20	2	71384.00	68	3.44	0.00	49059.13	41.00	0.24
06-064	35	2	116995.10	6	3.36	2	59603.00	80	5.91	0.00	-57392.10	74.00	2.55

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-065	36	3	35878.61	35	3.02	2	74256.00	138	3.03	-1.00	38377.39	103.00	0.01
06-066	60	1	41877.48	1	2.73	2	187744.00	69	4.08	1.00	145866.52	68.00	1.35
06-067	44	2	118240.89	6	7.75	2	39823.00	99	2.17	0.00	-78417.89	93.00	-5.58
06-068	66	2	100090.50	95	2.55	2	111690.00	166	1.91	0.00	11599.50	71.00	-0.64
06-069	37	2	23581.77	8	12.21	2	91317.00	107	1.92	0.00	67735.23	99.00	-10.29
06-070	76	2	193482.80	14	2.75	2	244546.00	35	3.10	0.00	51063.20	21.00	0.35
06-071	81	1	75525.40	0	5.02	3	77753.00	9	2.64	2.00	2227.60	9.00	-2.38
06-072	78	2	164658.40	17	2.75	2	14858.00	82	2.30	0.00	-149800.40	65.00	-0.45
06-073	97	2	30429.33	4	3.67	2	64324.00	38	3.32	0.00	33894.68	34.00	-0.35
06-074	79	1	148084.80	2	3.84	2	127354.00	31	3.02	1.00	-20730.80	29.00	-0.82
06-075	86	2	68936.15	2	4.64	1	23502.00	25	2.61	-1.00	-45434.15	23.00	-2.03
06-076	96	1	13557.68	78	2.42	3	72447.00	203	3.00	2.00	58889.32	125.00	0.58
06-077	68	2	89940.90	10	1.97	2	22463.00	53	2.24	0.00	-67477.90	43.00	0.27
06-078	87	2	215625.00	27	3.96	2	93362.00	56	4.97	0.00	-122263.00	29.00	1.01
06-079	93	2	27268.03	97	3.41	2	50690.00	100	3.33	0.00	23421.97	3.00	-0.08
06-080	69	1	121453.10	15	2.53	2	70697.00	31	4.09	1.00	-50756.10	16.00	1.56
06-081	98	2	.	40	2.59	2	125850.00	84	2.65	0.00	.	44.00	0.06
06-082	71	2	113748.90	54	3.73	2	65793.00	41	3.10	0.00	-47955.90	-13.00	-0.63
06-083	99	2	74747.60	20	2.38	2	99896.00	43	1.73	0.00	25148.40	23.00	-0.65
06-084	94	2	113438.20	5	3.51	2	37763.00	69	4.30	0.00	-75675.20	64.00	0.79
06-085	90	2	139809.04	6	4.70	2	43309.00	20	3.21	0.00	-96500.04	14.00	-1.49
06-086	95	3	259476.94	33	3.05	3	182168.00	109	2.21	0.00	-77308.94	76.00	-0.84
06-087	73	2	32933.76	0	6.12	3	127270.00	70	3.67	1.00	94336.24	70.00	-2.45
06-088	70	2	77004.29	2	3.35	2	78537.00	54	2.53	0.00	1532.71	52.00	-0.82
06-089	77	2	35170.11	91	3.36	2	68829.00	144	18.22	0.00	33658.89	53.00	14.86
06-090	85	2	105882.65	2	3.98	2	150274.00	28	5.86	0.00	44391.35	26.00	1.88
06-091	83	2	99698.35	15	3.84	2	59323.00	44	3.92	0.00	-40375.35	29.00	0.08
06-092	82	2	130182.97	1	3.85	2	129916.00	64	3.84	0.00	-266.97	63.00	-0.01
06-093	84	2	119957.97	0	4.71	2	144297.00	32	3.03	0.00	24339.03	32.00	-1.68
06-094	91	2	43791.30	6	1.67	3	92597.00	51	1.84	1.00	48805.70	45.00	0.17
06-095	88	2	70966.10	25	2.60	3	84762.00	70	2.95	1.00	13795.90	45.00	0.35
06-096	72	2	49545.21	147	1.63	2	97589.00	169	1.36	0.00	48043.79	22.00	-0.27
06-097	89	2	43826.72	20	3.90	2	18818.00	62	3.42	0.00	-25008.72	42.00	-0.48

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-098	100	2	121337.85	46	2.66	2	76217.00	77	2.61	0.00	-45120.85	31.00	-0.05
06-099	92	1	62622.10	29	2.51	2	60043.00	74	2.85	1.00	-2579.10	45.00	0.34
06-100	80	1	24585.52	0	4.69	1	32626.00	9	48.11	0.00	8040.48	9.00	43.42
06-101	75	3	159823.60	18	2.33	2	234470.00	25	1.79	-1.00	74646.40	7.00	-0.54
06-102	110	2	58540.62	19	2.75	3	208191.00	16	2.65	1.00	149650.38	-3.00	-0.10
06-103	109	2	100709.80	11	2.90	2	127854.00	63	2.50	0.00	27144.20	52.00	-0.40
06-104	116	1	61095.65	22	3.54	2	80738.00	69	8.69	1.00	19642.35	47.00	5.15
06-105	118	1	7857.66	19	2.73	3	340752.00	165	1.55	2.00	332894.34	146.00	-1.18
06-106	133	2	63716.12	26	3.24	1	38345.00	44	4.98	-1.00	-25371.12	18.00	1.74
06-107	126	1	15389.23	12	3.10	2	61559.00	26	2.94	1.00	46169.77	14.00	-0.16
06-108	112	1	229781.69	42	2.16	2	95766.00	41	2.35	1.00	-134015.69	-1.00	0.19
06-109	120	1	125440.95	6	2.10	1	46720.00	28	2.47	0.00	-78720.95	22.00	0.37
06-110	124	1	22452.46	28	2.78	2	48264.00	60	7.48	1.00	25811.54	32.00	4.70
06-111	104	2	37103.63	34	3.58	2	95470.00	58	4.06	0.00	58366.37	24.00	0.48
06-112	113	2	22620.27	100	2.37	2	75646.00	121	6.83	0.00	53025.73	21.00	4.46
06-113	122	2	115737.6	7	2.49	2	83356.00	8	5.32	0.00	-32381.60	1.00	2.83
06-114	121	1	28124.15	2	2.94	2	122574.00	99	2.14	1.00	94449.85	97.00	-0.80
06-115	117	3	115887.36	78	2.22	3	120747.00	272	3.63	0.00	4859.64	194.00	1.41
06-116	115	2	126538.50	35	3.10	3	103594.00	74	3.71	1.00	-22944.50	39.00	0.61
06-117	129	2	73213.70	0	5.42	2	135912.00	40	7.79	0.00	62698.30	40.00	2.37
06-118	130	2	75831.40	12	3.07	4	183342.00	26	1.97	2.00	107510.60	14.00	-1.10
06-119	127	2	98560.40	19	3.09	2	63148.00	43	2.59	0.00	-35412.40	24.00	-0.50
06-120	132	2	102090.37	44	3.00	1	38551.00	69	3.78	-1.00	-63539.37	25.00	0.78
06-121	108	2	66974.80	7	2.91	3	128301.00	27	6.17	1.00	61326.20	20.00	3.26
06-122	106	2	141818.70	29	2.98	2	96933.00	35	3.32	0.00	-44885.70	6.00	0.34
06-123	105	2	84739.77	20	3.78	3	133417.00	17	3.02	1.00	48677.23	-3.00	-0.76
06-124	101	.	.	9	2.89	2	77832.00	74	1.85	.	.	65.00	-1.04
06-125	119	2	23261.41	6	4.34	2	99751.00	145	3.29	0.00	76489.59	139.00	-1.05
06-126	111	2	47581.99	10	3.15	2	79880.00	111	5.12	0.00	32298.01	101.00	1.97
06-127	123	1	16303.57	29	2.06	3	84801.00	208	1.67	2.00	68497.43	179.00	-0.39
06-128	125	2	20930.25	15	3.51	2	32358.00	72	3.82	0.00	11427.75	57.00	0.31
06-129	128	3	149116.86	13	2.37	3	259273.00	73	2.11	0.00	110156.14	60.00	-0.26
06-130	107	3	141578.13	72	0.98	4	244270.00	203	1.10	1.00	102691.87	131.00	0.12

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-131	102	2	143185.10	13	3.57	2	193093.00	75	3.67	0.00	49907.90	62.00	0.10
06-132	131	3	287341.30	98	0.99	2	125942.00	203	0.80	-1.00	-161399.3	105.00	-0.19
06-133	114	1	41795.69	15	3.60	2	72145.00	15	2.73	1.00	30349.31	0.00	-0.87
06-134	103	1	8607.92	3	3.93	2	9413.90	54	3.61	1.00	805.98	51.00	-0.32
06-135	74	2	90757.20	7	2.69	3	322651.00	58	2.01	1.00	231893.80	51.00	-0.68
06-136	147	1	119134.90	79	3.27	2	.	118	2.31	1.00	.	39.00	-0.96
06-137	164	2	115594.40	12	3.48	2	180090.00	65	3.47	0.00	64495.60	53.00	-0.01
06-138	146	1	46101.43	2	2.46	1	93716.00	52	1.92	0.00	47614.57	50.00	-0.54
06-139	162	2	63783.80	13	2.96	2	59858.00	57	1.99	0.00	-3925.80	44.00	-0.97
06-140	144	2	109535.10	13	3.04	2	50150.00	31	7.19	0.00	-59385.10	18.00	4.15
06-141	163	2	92233.45	58	3.51	3	51368.00	65	5.81	1.00	-40865.45	7.00	2.30
06-142	148	2	122423.50	16	2.68	2	49654.00	151	1.41	0.00	-72769.50	135.00	-1.27
06-143	158	1	20622.43	9	3.06	3	109126.00	44	2.83	2.00	88503.58	35.00	-0.23
06-144	140	2	66232.68	9	2.55	2	82472.00	21	2.41	0.00	16239.32	12.00	-0.14
06-145	145	2	109038.60	2	1.13	2	128730.00	17	1.42	0.00	19691.40	15.00	0.29
06-146	136	1	18817.75	3	3.18	2	54652.00	70	3.25	1.00	35834.25	67.00	0.07
06-147	155	2	41599.04	5	2.21	2	51302.00	70	1.46	0.00	9702.96	65.00	-0.75
06-148	142	1	99661.80	119	2.93	2	45041.00	62	3.37	1.00	-54620.80	-57.00	0.44
06-149	152	1	25945.74	25	4.72	2	56554.00	47	4.22	1.00	30608.26	22.00	-0.50
06-150	159	2	52861.32	28	4.73	2	32445.00	172	5.25	0.00	-20416.32	144.00	0.52
06-151	138	2	104020.60	0	3.44	2	50071.00	60	5.57	0.00	-53949.60	60.00	2.13
06-152	139	1	61968.33	3	7.23	2	41475.00	81	5.50	1.00	-20493.33	78.00	-1.73
06-153	156	2	106110.66	39	3.25	2	60898.00	57	2.22	0.00	-45212.66	18.00	-1.03
06-154	135	2	143455.90	3	3.11	2	100337.00	13	2.60	0.00	-43118.90	10.00	-0.51
06-155	137	2	52362.67	13	3.16	2	83848.00	24	5.27	0.00	31485.33	11.00	2.11
06-156	166	2	84462.10	4	4.09	2	138010.00	18	3.93	0.00	53547.90	14.00	-0.16
06-157	165	1	38568.43	8	3.50	2	56315.00	5	2.40	1.00	17746.57	-3.00	-1.10
06-158	149	1	43889.60	16	6.67	2	40352.00	96	3.56	1.00	-3537.60	80.00	-3.11
06-159	151	1	76124.40	0	4.99	2	72209.00	60	8.86	1.00	-3915.40	60.00	3.87
06-160	143	2	37690.77	16	3.49	2	120968.00	53	3.98	0.00	83277.23	37.00	0.49
06-161	161	2	51996.78	9	3.61	2	109735.00	98	2.75	0.00	57738.22	89.00	-0.86
06-162	160	1	32651.04	0	3.32	3	226979.00	33	2.93	2.00	194327.96	33.00	-0.39
06-163	141	3	157920.55	45	1.61	4	341521.00	107	1.44	1.00	183600.45	62.00	-0.17

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-164	153	4	144616.02	26	1.60	5	123362.00	29	1.84	1.00	-21254.02	3.00	0.24
06-165	134	1	149573.34	4	3.19	2	123671.00	91	7.53	1.00	-25902.34	87.00	4.34
06-166	154	1	22430.31	9	3.24	2	83635.00	55	3.26	1.00	61204.69	46.00	0.02
06-167	157	2	180469.40	4	2.56	3	191046.00	172	1.44	1.00	10576.60	168.00	-1.12
06-168	150	1	36165.86	20	3.40	2	117671.00	52	9.29	1.00	81505.14	32.00	5.89
06-169	221	3	156161.32	7	2.26	2	81826.00	23	3.32	-1.00	-74335.32	16.00	1.06
06-170	218	1	69451.80	9	4.95	2	82013.00	68	3.20	1.00	12561.20	59.00	-1.75
06-171	202	2	109440.10	11	4.41	2	72077.00	54	2.87	0.00	-37363.10	43.00	-1.54
06-172	201	1	86170.30	7	3.87	1	29090.00	61	2.67	0.00	-57080.30	54.00	-1.20
06-173	223	2	131775.90	12	3.44	2	113852.00	50	2.75	0.00	-17923.90	38.00	-0.69
06-174	203	2	88220.43	3	3.06	2	135034.00	35	4.13	0.00	46813.57	32.00	1.07
06-175	216	2	28575.20	14	2.72	2	74996.00	20	5.39	0.00	46420.81	6.00	2.67
06-176	211	2	106155.10	2	6.03	2	69885.00	33	3.36	0.00	-36270.10	31.00	-2.67
06-177	210	2	80019.04	35	3.04	2	79683.00	16	3.23	0.00	-336.03	-19.00	0.19
06-178	215	1	72483.20	14	4.53	2	62434.00	11	3.99	1.00	-10049.20	-3.00	-0.54
06-179	200	1	86249.70	3	3.52	2	128662.00	34	2.86	1.00	42412.30	31.00	-0.66
06-180	209	1	23426.23	41	3.00	2	41597.00	85	2.93	1.00	18170.77	44.00	-0.07
06-181	212	2	103102.80	4	4.60	2	95800.00	97	3.10	0.00	-7302.80	93.00	-1.50
06-182	206	2	102579.40	31	3.32	2	60209.00	109	7.34	0.00	-42370.40	78.00	4.02
06-183	225	2	135668.40	3	13.65	3	98880.00	46	4.75	1.00	-36788.40	43.00	-8.90
06-184	229	2	64553.06	23	4.04	2	.	71	4.00	0.00	.	48.00	-0.04
06-185	204	2	119816.94	9	5.83	2	115804.00	118	2.47	0.00	-4012.94	109.00	-3.36
06-186	222	3	124469.55	1	2.85	2	130827.00	11	2.21	-1.00	6357.45	10.00	-0.64
06-187	219	1	19162.89	55	3.72	2	50508.00	107	2.42	1.00	31345.11	52.00	-1.30
06-188	205	1	90225.88	9	3.70	2	176869.00	32	2.51	1.00	86643.12	23.00	-1.19
06-189	213	3	173024.36	41	2.87	3	170920.00	25	2.37	0.00	-2104.36	-16.00	-0.50
06-190	208	2	61761.37	1	3.02	1	25198.00	44	13.47	-1.00	-36563.37	43.00	10.45
06-191	224	2	212240.00	4	1.43	2	167198.00	25	1.47	0.00	-45042.00	21.00	0.04
06-192	226	1	34353.61	13	4.78	2	74544.00	25	3.89	1.00	40190.39	12.00	-0.89
06-193	217	3	205775.30	45	7.06	2	127010.00	66	3.35	-1.00	-78765.30	21.00	-3.71
06-194	227	2	131803.27	30	3.60	2	99491.00	71	3.57	0.00	-32312.27	41.00	-0.03
06-195	207	2	113306.90	45	3.38	2	85351.00	84	1.88	0.00	-27955.90	39.00	-1.50
06-196	220	2	190122.80	4	2.82	2	205203.00	52	2.24	0.00	15080.20	48.00	-0.58

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-197	214	3	112104.42	22	2.78	2	21415.00	126	1.85	-1.00	-90689.42	104.00	-0.93
06-198	228	2	40143.01	18	4.19	3	99171.00	92	2.50	1.00	59027.99	74.00	-1.69
06-199	180	2	88580.60	22	4.77	2	57339.00	27	4.81	0.00	-31241.60	5.00	0.04
06-200	184	2	50867.05	3	3.93	2	46580.00	35	3.02	0.00	-4287.05	32.00	-0.91
06-201	188	3	177170.40	20	4.10	2	39644.00	60	8.28	-1.00	-137526.4	40.00	4.18
06-202	186	1	205719.24	7	3.97	2	29926.00	58	1.89	1.00	-175793.24	51.00	-2.08
06-203	194	2	43668.89	86	3.53	2	51941.00	166	1.47	0.00	8272.11	80.00	-2.06
06-204	179	2	129423.35	4	3.83	2	46524.00	20	1.95	0.00	-82899.35	16.00	-1.88
06-205	174	2	85484.10	66	1.49	2	141918.00	111	1.47	0.00	56433.90	45.00	-0.02
06-206	170	2	70327.70	4	3.24	2	24921.00	44	26.35	0.00	-45406.70	40.00	23.11
06-207	185	2	14622.54	0	5.21	1	20605.00	23	3.29	-1.00	5982.46	23.00	-1.92
06-208	196	1	569319.24	45	3.64	1	28199.00	56	3.59	0.00	-541120.2	11.00	-0.05
06-209	182	2	105336.01	5	3.73	2	15801.00	33	3.24	0.00	-89535.01	28.00	-0.49
06-210	171	2	72034.65	2	3.58	2	37976.00	57	2.09	0.00	-34058.65	55.00	-1.49
06-211	183	1	72709.80	13	4.45	2	70949.00	60	2.65	1.00	-1760.80	47.00	-1.80
06-212	173	2	53103.69	5	3.00	2	98845.00	45	3.24	0.00	45741.31	40.00	0.24
06-213	181	1	19736.90	9	5.85	2	77414.00	119	2.87	1.00	57677.10	110.00	-2.98
06-214	191	2	249457.00	3	3.67	3	335512.00	144	1.23	1.00	86055.00	141.00	-2.44
06-215	187	2	152235.40	5	4.65	2	112980.00	9	5.96	0.00	-39255.40	4.00	1.31
06-216	199	2	85014.89	5	4.63	1	47759.00	16	4.62	-1.00	-37255.89	11.00	-0.01
06-217	197	2	63565.69	6	6.24	2	44872.00	27	5.20	0.00	-18693.69	21.00	-1.04
06-218	193	2	30508.50	3	6.12	2	115577.00	21	3.42	0.00	85068.50	18.00	-2.70
06-219	189	2	86223.20	0	4.23	2	78701.00	152	4.21	0.00	-7522.20	152.00	-0.02
06-220	178	2	136920.10	10	2.75	2	145824.00	135	2.39	0.00	8903.90	125.00	-0.36
06-221	168	2	121050.71	12	2.72	3	124827.00	59	2.56	1.00	3776.29	47.00	-0.16
06-223	195	1	35967.29	14	3.60	2	162038.00	14	4.05	1.00	126070.71	0.00	0.45
06-224	167	3	132997.00	1	2.25	2	116673.00	95	1.72	-1.00	-16324.00	94.00	-0.53
06-225	175	2	89225.24	9	1.33	3	131141.00	87	1.31	1.00	41915.76	78.00	-0.02
06-226	169	1	79118.90	9	3.06	2	49115.00	112	3.49	1.00	-30003.90	103.00	0.43
06-227	177	1	134986.80	24	1.22	2	115690.00	39	1.11	1.00	-19296.80	15.00	-0.11
06-228	192	2	27684.14	2	4.15	2	113838.00	37	2.84	0.00	86153.86	35.00	-1.31
06-229	176	5	85343.45	91	1.84	4	103529.00	179	1.33	-1.00	18185.55	88.00	-0.51
06-230	198	4	104360.29	74	2.65	2	19336.00	57	1.46	-2.00	-85024.29	-17.00	-1.19

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-231	190	3	197248.00	30	3.07	3	20362.00	138	1.00	0.00	-176886.0	108.00	-2.07
06-232	172	1	30851.45	20	3.07	2	55849.00	78	2.33	1.00	24997.55	58.00	-0.74
06-233	237	2	157988.60	8	3.69	2	45056.00	9	23.97	0.00	-112932.6	1.00	20.28
06-234	258	2	36755.08	4	4.26	2	44827.00	33	3.42	0.00	8071.92	29.00	-0.84
06-235	265	2	38823.94	6	4.26	2	28703.00	2	5.63	0.00	-10120.94	-4.00	1.37
06-236	249	2	94843.90	10	3.41	2	66825.00	28	1.71	0.00	-28018.90	18.00	-1.70
06-237	236	2	83810.10	13	2.98	2	77279.00	83	2.30	0.00	-6531.10	70.00	-0.68
06-238	256	2	75854.00	74	2.65	2	106006.00	44	1.92	0.00	30152.00	-30.00	-0.73
06-239	238	1	5780.56	12	4.75	1	2230.40	19	28.44	0.00	-3550.16	7.00	23.69
06-240	239	2	101595.60	19	3.71	2	55514.00	105	16.68	0.00	-46081.60	86.00	12.97
06-241	230	1	29588.82	6	3.44	2	73571.00	3	4.58	1.00	43982.18	-3.00	1.14
06-242	232	1	34426.49	5	4.22	2	96512.00	13	2.91	1.00	62085.51	8.00	-1.31
06-243	233	3	121157.35	30	3.19	2	89429.00	34	2.86	-1.00	-31728.35	4.00	-0.33
06-244	251	1	38887.49	24	3.35	2	57632.00	23	3.44	1.00	18744.51	-1.00	0.09
06-245	259	2	42884.76	91	3.08	2	121397.00	35	1.88	0.00	78512.24	-56.00	-1.20
06-246	243	3	75612.61	0	2.36	2	49779.00	16	2.89	-1.00	-25833.61	16.00	0.53
06-247	231	1	19105.26	25	2.66	2	38106.00	71	7.24	1.00	19000.74	46.00	4.58
06-248	252	2	111356.90	17	2.92	2	105215.00	16	2.95	0.00	-6141.90	-1.00	0.03
06-249	262	2	135851.00	11	5.03	2	39748.00	29	7.16	0.00	-96103.00	18.00	2.13
06-250	253	3	77798.07	18	2.97	2	13344.00	32	8.76	-1.00	-64454.07	14.00	5.79
06-251	246	2	46462.16	6	3.41	2	60269.00	49	3.37	0.00	13806.84	43.00	-0.04
06-252	241	2	66130.10	37	2.80	2	15733.00	113	5.50	0.00	-50397.10	76.00	2.70
06-253	254	2	109625.10	9	3.18	2	42303.00	11	24.90	0.00	-67322.10	2.00	21.72
06-254	257	1	50152.05	71	5.70	1	32048.00	126	5.56	0.00	-18104.05	55.00	-0.14
06-255	267	2	59251.30	36	2.68	2	77888.00	40	9.43	0.00	18636.70	4.00	6.75
06-256	242	2	75619.42	1	4.28	2	51705.00	19	24.14	0.00	-23914.42	18.00	19.86
06-257	247	2	104259.64	50	2.34	1	8348.00	108	4.31	-1.00	-95911.64	58.00	1.97
06-258	255	2	64935.50	8	3.54	2	47930.00	20	9.07	0.00	-17005.50	12.00	5.53
06-259	260	2	8026.53	17	3.32	3	121080.00	44	3.41	1.00	113053.47	27.00	0.09
06-260	234	2	29166.63	25	2.43	2	35554.00	141	2.20	0.00	6387.37	116.00	-0.23
06-261	264	1	76933.20	7	2.66	1	18551.00	26	4.81	0.00	-58382.20	19.00	2.15
06-262	266	2	135370.00	5	4.12	2	54942.00	73	3.26	0.00	-80428.00	68.00	-0.86
06-263	235	1	81672.70	6	3.17	1	39058.00	28	2.31	0.00	-42614.70	22.00	-0.86

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-264	268	3	158490.60	5	3.59	3	98908.00	13	4.82	0.00	-59582.60	8.00	1.23
06-265	250	3	141852.11	14	2.63	2	51276.00	34	4.12	-1.00	-90576.11	20.00	1.49
06-266	334	2	148224.40	25	2.96	3	177599.00	122	1.43	1.00	29374.60	97.00	-1.53
06-267	330	2	80150.54	35	2.57	2	43691.00	68	2.63	0.00	-36459.54	33.00	0.06
06-268	314	2	75638.92	27	2.34	2	56379.00	59	1.69	0.00	-19259.92	32.00	-0.65
06-269	333	2	6391.33	49	2.67	2	48059.00	77	2.61	0.00	41667.67	28.00	-0.06
06-270	328	2	51758.06	14	3.64	2	138297.00	5	4.39	0.00	86538.94	-9.00	0.75
06-271	322	2	70193.61	7	3.67	2	25568.00	33	4.55	0.00	-44625.61	26.00	0.88
06-272	307	2	100814.84	3	4.85	2	53703.00	13	4.45	0.00	-47111.84	10.00	-0.40
06-273	336	2	74221.87	6	2.84	2	18200.00	9	36.15	0.00	-56021.87	3.00	33.31
06-274	337	2	65481.19	6	3.07	1	33347.00	32	2.66	-1.00	-32134.19	26.00	-0.41
06-275	339	2	36468.40	0	4.25	1	18496.00	2	3.63	-1.00	-17972.40	2.00	-0.62
06-276	315	2	107216.00	58	2.94	3	138720.00	22	3.02	1.00	31504.00	-36.00	0.08
06-277	326	3	119651.80	15	3.66	2	128747.00	43	2.79	-1.00	9095.20	28.00	-0.87
06-278	309	2	39088.60	0	6.75	2	59554.00	11	4.00	0.00	20465.40	11.00	-2.75
06-279	325	3	43353.74	34	3.77	3	68141.00	31	2.34	0.00	24787.26	-3.00	-1.43
06-280	327	2	123282.50	0	4.00	3	153046.00	16	2.31	1.00	29763.50	16.00	-1.69
06-281	317	1	43379.21	18	3.50	2	15109.00	41	5.36	1.00	-28270.21	23.00	1.86
06-282	320	2	80914.10	23	2.80	2	44204.00	16	2.84	0.00	-36710.10	-7.00	0.04
06-283	319	2	63991.41	12	3.70	2	62629.00	56	4.19	0.00	-1362.41	44.00	0.49
06-284	323	2	237008.00	52	2.78	4	188368.00	133	3.36	2.00	-48640.00	81.00	0.58
06-285	335	2	45526.51	5	2.85	2	79357.00	8	62.70	0.00	33830.49	3.00	59.85
06-286	312	2	93506.84	17	6.60	2	58641.00	63	3.31	0.00	-34865.84	46.00	-3.29
06-287	340	1	49606.70	16	4.39	2	84443.00	54	4.37	1.00	34836.30	38.00	-0.02
06-288	324	2	160950.50	23	6.17	2	79333.00	15	3.84	0.00	-81617.50	-8.00	-2.33
06-289	329	1	117794.90	8	2.79	2	101095.00	5	1.90	1.00	-16699.90	-3.00	-0.89
06-290	308	2	133100.10	22	2.95	3	64117.00	14	1.72	1.00	-68983.10	-8.00	-1.23
06-291	311	2	65220.07	14	3.61	2	49349.00	17	3.44	0.00	-15871.07	3.00	-0.17
06-292	313	2	142618.90	0	4.64	2	178851.00	14	2.37	0.00	36232.10	14.00	-2.27
06-293	310	3	61675.42	62	2.20	3	150785.00	86	1.49	0.00	89109.58	24.00	-0.71
06-294	332	2	69657.67	10	3.84	2	124471.00	63	3.38	0.00	54813.33	53.00	-0.46
06-295	331	2	45322.00	8	3.33	2	65628.00	46	3.09	0.00	20306.00	38.00	-0.24
06-296	316	1	31238.74	8	2.71	2	108628.00	10	1.88	1.00	77389.26	2.00	-0.83

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-297	318	1	9548.07	0	4.57	2	18797.00	27	3.26	1.00	9248.93	27.00	-1.31
06-298	338	2	146485.20	19	2.51	2	139808.00	69	3.23	0.00	-6677.20	50.00	0.72
06-299	321	2	164918.15	13	3.40	2	109994.00	9	2.60	0.00	-54924.15	-4.00	-0.80
06-300	296	2	20789.56	6	3.34	1	25920.00	49	2.33	-1.00	5130.45	43.00	-1.01
06-301	301	2	57468.43	9	5.04	2	68296.00	17	6.49	0.00	10827.57	8.00	1.45
06-302	294	4	68241.29	15	3.53	3	86385.00	70	7.52	-1.00	18143.71	55.00	3.99
06-303	292	1	54992.10	33	3.21	2	30196.00	58	2.15	1.00	-24796.10	25.00	-1.06
06-304	289	2	124286.60	3	7.48	2	107979.00	2	4.37	0.00	-16307.60	-1.00	-3.11
06-305	263	2	73930.49	6	2.47	3	129008.00	5	19.06	1.00	55077.51	-1.00	16.59
06-306	290	1	19483.06	41	2.52	1	15179.00	67	4.76	0.00	-4304.06	26.00	2.24
06-307	277	2	13813.57	19	3.78	2	35869.00	56	2.47	0.00	22055.43	37.00	-1.31
06-308	304	2	55802.16	9	3.20	2	30601.00	79	10.36	0.00	-25201.16	70.00	7.16
06-309	281	2	134286.17	13	4.07	2	88991.00	22	3.46	0.00	-45295.17	9.00	-0.61
06-310	293	1	61305.39	19	4.50	2	152601.00	7	12.92	1.00	91295.61	-12.00	8.42
06-311	269	1	88206.50	29	3.03	2	225679.00	11	3.29	1.00	137472.50	-18.00	0.26
06-312	240	2	133186.30	95	1.75	3	73429.00	179	1.22	1.00	-59757.30	84.00	-0.53
06-313	288	2	55859.38	30	3.56	2	46310.00	43	5.74	0.00	-9549.38	13.00	2.18
06-314	274	2	134312.60	27	3.12	2	151980.00	48	4.33	0.00	17667.40	21.00	1.21
06-315	272	2	40447.86	4	2.92	2	83711.00	16	2.92	0.00	43263.14	12.00	0.00
06-316	271	1	23336.21	36	3.58	2	27955.00	48	4.09	1.00	4618.79	12.00	0.51
06-317	306	2	155383.10	14	1.83	3	162426.00	108	2.18	1.00	7042.90	94.00	0.35
06-318	248	3	117956.14	47	2.14	3	208247.00	169	1.81	0.00	90290.86	122.00	-0.33
06-319	287	2	81328.00	113	3.25	2	66992.00	81	5.64	0.00	-14336.00	-32.00	2.39
06-320	280	1	44054.62	12	4.32	2	38787.00	77	15.52	1.00	-5267.62	65.00	11.20
06-321	285	3	67430.01	30	1.66	3	159777.00	20	1.47	0.00	92346.99	-10.00	-0.19
06-322	283	2	91821.80	9	5.26	3	107105.00	20	2.16	1.00	15283.20	11.00	-3.10
06-323	295	2	145760.00	19	3.17	2	189342.00	13	12.58	0.00	43582.00	-6.00	9.41
06-324	245	3	46749.91	11	3.15	3	75280.00	18	2.90	0.00	28530.09	7.00	-0.25
06-325	278	2	55909.87	21	5.12	2	116176.00	68	23.31	0.00	60266.13	47.00	18.19
06-326	291	3	147199.37	41	3.05	2	175496.00	96	3.35	-1.00	28296.63	55.00	0.30
06-327	300	3	138307.70	101	1.12	1	15182.00	115	0.94	-2.00	-123125.7	14.00	-0.18
06-328	270	2	43650.44	5	3.74	2	36084.00	42	3.70	0.00	-7566.44	37.00	-0.04
06-329	305	1	98148.60	36	2.59	2	65383.00	66	3.76	1.00	-32765.60	30.00	1.17

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-330	303	5	187624.83	18	1.49	.	.	59	2.80	.	.	41.00	1.31
06-331	273	3	113478.01	14	2.84	2	100801.00	45	1.66	-1.00	-12677.01	31.00	-1.18
06-332	261	2	141660.10	12	4.31	2	189683.00	10	2.85	0.00	48022.90	-2.00	-1.46
06-333	275	2	74435.01	16	3.87	2	127208.00	42	3.84	0.00	52772.99	26.00	-0.03
06-334	348	2	159826.90	53	3.25	2	51497.00	84	2.20	0.00	-108329.9	31.00	-1.05
06-335	366	1	80125.50	51	1.69	2	93393.00	50	1.79	1.00	13267.50	-1.00	0.10
06-336	365	3	150517.48	55	2.21	2	86275.00	68	1.81	-1.00	-64242.48	13.00	-0.40
06-337	350	2	145399.60	11	3.71	2	98671.00	7	10.81	0.00	-46728.60	-4.00	7.10
06-338	345	2	81705.30	7	3.35	2	67172.00	12	2.43	0.00	-14533.30	5.00	-0.92
06-339	359	2	320848.54	14	1.60	2	.	18	1.64	0.00	.	4.00	0.04
06-340	347	3	137273.21	11	2.45	2	75935.00	92	4.88	-1.00	-61338.21	81.00	2.43
06-341	361	1	49787.20	16	3.16	2	21238.00	37	2.43	1.00	-28549.20	21.00	-0.73
06-342	346	2	83911.67	1	4.31	2	54479.00	8	37.03	0.00	-29432.67	7.00	32.72
06-343	372	2	107180.60	7	2.69	2	38863.00	7	2.03	0.00	-68317.60	0.00	-0.66
06-344	356	2	61082.43	19	2.76	2	54180.00	26	2.75	0.00	-6902.43	7.00	-0.01
06-345	362	2	89132.41	7	3.69	2	82841.00	67	3.99	0.00	-6291.40	60.00	0.30
06-346	369	2	198645.71	12	4.26	2	131615.00	13	2.00	0.00	-67030.71	1.00	-2.26
06-347	352	3	164177.40	41	1.49	3	225130.00	102	1.49	0.00	60952.60	61.00	0.00
06-348	374	1	82244.50	3	4.09	2	93727.00	12	3.22	1.00	11482.50	9.00	-0.87
06-349	373	1	109409.10	20	2.77	1	23310.00	7	2.18	0.00	-86099.10	-13.00	-0.59
06-350	355	2	126446.80	6	2.57	2	60578.00	32	2.67	0.00	-65868.80	26.00	0.10
06-351	349	2	63638.66	15	3.96	3	129360.00	46	1.59	1.00	65721.34	31.00	-2.37
06-352	353	2	104932.98	11	3.52	1	29986.00	37	2.86	-1.00	-74946.98	26.00	-0.66
06-353	343	2	178236.20	20	3.00	3	159667.00	62	2.37	1.00	-18569.20	42.00	-0.63
06-354	360	1	62695.40	32	3.43	2	49354.00	20	2.63	1.00	-13341.40	-12.00	-0.80
06-355	354	1	48296.10	32	3.09	2	108880.00	59	3.77	1.00	60583.90	27.00	0.68
06-356	351	2	115953.40	12	3.95	2	147228.00	4	3.78	0.00	31274.60	-8.00	-0.17
06-357	367	2	85759.90	11	2.90	2	50569.00	26	2.39	0.00	-35190.90	15.00	-0.51
06-358	370	3	82158.80	14	2.33	3	35788.00	131	0.99	0.00	-46370.80	117.00	-1.34
06-359	341	2	109292.50	29	1.56	3	167200.00	160	1.12	1.00	57907.50	131.00	-0.44
06-360	358	2	82863.50	6	2.53	2	50769.00	54	2.25	0.00	-32094.50	48.00	-0.28
06-361	371	3	191215.80	51	1.56	3	229464.00	89	1.32	0.00	38248.20	38.00	-0.24
06-362	344	1	90345.98	10	3.83	2	86449.00	16	2.42	1.00	-3896.98	6.00	-1.41

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-363	342	2	117261.30	11	2.88	2	50810.00	20	3.38	0.00	-66451.30	9.00	0.50
06-364	363	2	126811.80	11	3.00	2	56933.00	16	3.31	0.00	-69878.80	5.00	0.31
06-365	364	2	119463.30	15	4.69	2	144226.00	8	2.91	0.00	24762.70	-7.00	-1.78
06-366	357	2	114235.00	22	4.12	2	206966.00	8	4.72	0.00	92731.00	-14.00	0.60
06-367	298	4	208113.10	109	2.08	3	189482.00	151	2.11	-1.00	-18631.10	42.00	0.03
06-368	279	1	53788.80	21	1.78	2	57719.00	26	1.85	1.00	3930.20	5.00	0.07
06-369	385	3	191273.30	5	4.02	2	61356.00	59	3.26	-1.00	-129917.3	54.00	-0.76
06-370	276	3	90723.59	29	4.34	3	59529.00	149	2.42	0.00	-31194.59	120.00	-1.92
06-371	384	2	135610.90	32	2.59	2	64729.00	20	2.48	0.00	-70881.90	-12.00	-0.11
06-372	388	2	49259.26	4	3.85	3	49259.00	52	5.74	1.00	-0.26	48.00	1.89
06-373	394	2	99479.66	13	2.43	2	67626.00	7	3.18	0.00	-31853.66	-6.00	0.75
06-374	284	3	81599.78	15	3.19	2	62452.00	35	2.77	-1.00	-19147.78	20.00	-0.42
06-375	380	3	115400.90	2	4.14	3	87085.00	43	3.10	0.00	-28315.90	41.00	-1.04
06-376	397	2	49336.60	13	3.35	2	49892.00	35	3.87	0.00	555.40	22.00	0.52
06-377	282	1	55905.88	19	1.93	2	31639.00	49	1.99	1.00	-24266.88	30.00	0.06
06-378	378	2	114294.80	4	4.81	2	154644.00	11	2.79	0.00	40349.20	7.00	-2.02
06-379	375	2	58527.10	12	3.11	2	26892.00	96	2.40	0.00	-31635.10	84.00	-0.71
06-380	400	2	49417.00	34	3.07	2	31383.00	21	50.16	0.00	-18034.00	-13.00	47.09
06-381	379	1	65910.20	30	3.20	2	86796.00	33	4.63	1.00	20885.80	3.00	1.43
06-382	386	2	142971.70	51	2.81	2	119986.00	67	3.79	0.00	-22985.70	16.00	0.98
06-383	286	1	44690.31	27	2.94	2	63295.00	38	3.98	1.00	18604.69	11.00	1.04
06-384	391	2	151538.00	13	4.25	2	84838.00	32	3.79	0.00	-66700.00	19.00	-0.46
06-385	377	2	136579.70	6	4.74	2	114310.00	120	4.34	0.00	-22269.70	114.00	-0.40
06-386	398	2	127792.50	6	3.01	2	45205.00	20	2.27	0.00	-82587.50	14.00	-0.74
06-387	390	2	67512.09	4	3.38	2	15297.00	20	3.82	0.00	-52215.09	16.00	0.44
06-388	299	2	111765.20	39	6.77	3	144890.00	76	4.42	1.00	33124.80	37.00	-2.35
06-389	399	2	72817.35	4	4.39	2	48661.00	26	11.99	0.00	-24156.35	22.00	7.60
06-390	382	2	90794.12	13	3.81	2	25339.00	14	7.27	0.00	-65455.12	1.00	3.46
06-391	376	2	25259.75	14	3.15	2	211793.00	30	1.93	0.00	186533.25	16.00	-1.22
06-392	383	3	178197.90	40	3.48	2	78301.00	54	5.29	-1.00	-99896.90	14.00	.
06-393	297	2	93313.39	14	3.36	2	148014.00	50	3.46	0.00	54700.61	36.00	0.10
06-394	393	2	54307.70	9	5.94	3	26269.00	77	4.22	1.00	-28038.70	68.00	-1.72
06-395	381	2	140570.48	54	2.91	2	31636.00	57	1.76	0.00	-108934.5	3.00	-1.15

Lab #	Tag #	SS 1	ASF 1	Peak 1	ET 1	SS 3	ASF 3	Peak 3	ET 3	SS 3-1	ASF 3-1	Peak 3-1	ET 3-1
06-396	389	3	205986.40	71	1.98	2	26483.00	95	1.67	-1.00	-179503.4	24.00	-0.31
06-397	396	2	144139.30	65	1.82	2	23274.00	41	1.77	0.00	-120865.3	-24.00	-0.05
06-398	387	3	233976.60	96	2.08	3	238667.00	132	1.87	0.00	4690.40	36.00	-0.21
06-399	302	3	183276.20	80	1.77	2	122181.00	45	2.51	-1.00	-61095.20	-35.00	0.74
06-400	395	4	140195.83	31	1.71	3	131079.00	102	1.19	-1.00	-9116.83	71.00	-0.52
06-401	392	2	152311.80	7	2.58	2	180766.00	58	3.07	0.00	28454.20	51.00	0.49

Appendix E Backgrounding Growth Data

Lab #	Tag #	SOT Wt (kg)	EOT Wt (kg)	Ship Wt (kg)	ADG (kg/day)	BF (mm)	REA (cm^2)
06-001	12	253.0	439	489	1.52	1.0	63.80
06-002	7	262.0	424	468	1.33	3.4	75.34
06-003	5	211.0	359	411	1.21	1.4	70.69
06-004	20	272.0	439	489	1.37	1.0	77.19
06-005	25	240.0	385	423	1.19	1.9	61.30
06-006	19	232.0	361	413	1.06	3.4	60.91
06-007	23	220.0	378	421	1.30	2.5	73.61
06-008	18	286.0	482	536	1.61	4.4	87.32
06-009	27	218.0	384	427	1.36	2.5	62.99
06-010	3	223.0	378	457	1.27	0.0	55.09
06-011	21	230.0	401	452	1.40	2.9	66.70
06-012	33	253.0	406	465	1.25	3.8	64.50
06-013	26	247.0	404	449	1.29	1.9	72.34
06-014	4	233.0	409	470	1.44	2.0	54.44
06-015	15	228.0	386	438	1.30	1.4	70.36
06-016	17	226.0	451	520	1.84	4.3	71.27
06-017	14	255.0	396	442	1.16	1.0	65.33
06-018	29	227.0	373	414	1.20	2.9	62.44
06-019	13	238.0	435	484	1.61	2.4	66.19
06-020	2	223.0	414	465	1.57	2.4	68.98
06-021	16	227.0	340	386	0.93	2.9	55.08
06-022	8	244.0	400	449	1.28	1.9	62.31
06-023	1	228.0	396	369	1.38	5.3	70.07
06-024	10	223.0	364	410	1.16	2.9	60.60
06-025	30	276.0	446	489	1.39	4.3	72.10
06-026	28	236.0	417	459	1.48	2.4	66.75
06-027	32	259.0	437	479	1.46	3.4	76.30
06-028	9	245.0	400	446	1.27	5.8	72.98
06-029	31	244.0	413	471	1.39	3.4	57.65
06-030	24	281.0	444	486	1.34	2.9	62.77
06-031	22	257.0	406	442	1.22	1.0	69.84
06-032	11	242.0	374	416	1.08	0.0	66.20
06-033	6	234.0	383	417	1.22	1.0	59.15
06-034	65	227.0	392	458	1.35	1.0	61.39
06-035	41	203.0	350	400	1.20	1.9	62.64
06-036	39	247.0	381	424	1.10	1.0	64.61
06-037	64	250.0	418	463	1.38	1.9	79.59
06-040	50	266.0	418	460	1.25	4.3	75.57
06-041	55	246.0	394	444	1.21	3.9	77.11
06-042	52	245.0	392	436	1.20	1.9	58.91
06-043	54	223.0	377	425	1.26	1.4	52.32

Lab #	Tag #	SOT Wt (kg)	EOT Wt (kg)	Ship Wt (kg)	ADG (kg/day)	BF (mm)	REA (cm^2)
06-044	51	202.0	342	384	1.15	0.0	47.05
06-045	48	258.0	423	472	1.35	1.6	63.48
06-046	40	253.0	418	450	1.35	2.4	78.86
06-047	45	229.0	415	468	1.52	1.0	76.93
06-048	43	282.0	480	534	1.62	2.9	80.16
06-049	49	257.0	438	478	1.48	1.0	55.90
06-050	61	233.0	393	439	1.31	1.0	61.17
06-051	59	247.0	361	412	0.93	0.0	66.78
06-052	38	221.0	359	392	1.13	2.4	71.23
06-053	46	256.0	355	387	0.81	1.0	66.06
06-054	53	202.0	309	359	0.88	2.0	54.72
06-055	67	218.0	363	406	1.19	2.0	59.57
06-056	56	249.0	387	429	1.13	1.9	59.60
06-057	47	264.0	437	477	1.42	1.5	82.61
06-058	57	262.0	446	492	1.51	6.7	58.17
06-059	63	219.0	363	417	1.18	1.0	60.84
06-060	58	251.0	400	453	1.22	6.3	59.40
06-061	34	240.0	409	469	1.39	1.0	72.52
06-062	42	218.0	343	390	1.02	1.0	53.33
06-063	62	256.0	436	497	1.48	2.9	62.28
06-064	35	246.0	412	465	1.36	1.0	60.31
06-065	36	268.0	449	510	1.48	1.4	73.88
06-066	60	188.0	311	354	1.01	1.9	50.33
06-067	44	212.0	382	436	1.39	1.0	62.22
06-068	66	273.0	409	447	1.11	2.4	59.96
06-069	37	272.0	411	445	1.14	1.0	77.30
06-070	76	251.0	451	512	1.64	2.0	63.42
06-071	81	265.0	407	443	1.17	3.8	65.35
06-072	78	238.0	367	400	1.06	1.0	54.34
06-073	97	259.0	376	429	0.97	1.4	71.74
06-074	79	253.0	387	420	1.11	1.0	56.84
06-075	86	231.0	362	412	1.08	1.9	56.79
06-076	96	215.0	367	415	1.26	1.6	59.46
06-077	68	241.0	396	429	1.27	1.0	60.38
06-078	87	236.0	361	399	1.03	2.9	68.25
06-079	93	227.0	358	393	1.08	2.4	69.40
06-080	69	247.0	384	429	1.12	1.4	71.57
06-081	98	249.0	397	439	1.22	2.9	85.85
06-082	71	249.0	402	453	1.25	1.4	67.20
06-083	99	238.0	363	394	1.03	1.6	66.23
06-084	94	244.0	366	408	1.01	2.5	62.73
06-085	90	266.0	383	431	0.97	2.4	67.75
06-086	95	246.0	362	395	0.96	1.9	63.83
06-087	73	288.0	417	448	1.06	4.3	74.18
06-088	70	207.0	342	385	1.11	0.0	60.91

Lab #	Tag #	SOT Wt (kg)	EOT Wt (kg)	Ship Wt (kg)	ADG (kg/day)	BF (mm)	REA (cm^2)
06-089	77	235.0	386	420	1.24	1.9	72.38
06-090	85	220.0	340	371	0.99	1.4	54.65
06-091	83	267.0	425	480	1.31	4.3	54.08
06-092	82	275.0	422	467	1.21	1.0	68.78
06-093	84	239.0	361	393	1.01	2.9	49.19
06-094	91	238.0	388	438	1.24	2.4	59.38
06-095	88	252.0	395	427	1.18	2.4	76.37
06-096	72	253.0	354	390	0.83	0.0	62.31
06-097	89	264.0	439	493	1.45	2.4	71.55
06-098	100	216.0	340	384	1.02	2.0	49.85
06-099	92	249.0	410	463	1.33	1.4	59.99
06-100	80	249.0	389	415	1.16	1.0	57.65
06-101	75	240.0	367	404	1.05	1.0	71.07
06-102	110	248.0	381	429	1.10	1.4	80.75
06-103	109	241.0	431	468	1.57	2.4	70.30
06-104	116	235.0	368	413	1.10	1.4	61.08
06-105	118	251.0	426	472	1.45	0.0	74.04
06-106	133	260.0	387	435	1.05	2.4	80.76
06-107	126	256.0	391	433	1.12	1.9	56.54
06-108	112	251.0	412	448	1.33	1.9	59.99
06-109	120	240.0	365	400	1.03	2.5	54.41
06-110	124	267.0	401	451	1.11	1.9	58.92
06-111	104	274.0	436	480	1.34	2.9	61.42
06-112	113	250.0	338	383	0.73	1.4	72.04
06-113	122	204.0	357	403	1.26	1.6	55.34
06-114	121	226.0	342	392	0.96	0.0	58.78
06-115	117	216.0	336	376	0.99	1.0	59.13
06-116	115	259.0	408	446	1.23	2.9	62.49
06-117	129	251.0	378	430	1.05	1.4	65.92
06-118	130	246.0	383	425	1.13	1.4	61.08
06-119	127	242.0	387	435	1.20	1.0	59.68
06-120	132	263.0	387	419	1.02	3.9	67.56
06-121	108	259.0	393	428	1.11	2.5	70.53
06-122	106	232.0	350	397	0.98	1.9	52.58
06-123	105	262.0	404	450	1.17	2.4	65.95
06-124	101	214.0	329	373	0.95	1.9	51.37
06-125	119	228.0	367	394	1.15	3.4	55.48
06-126	111	228.0	384	431	1.29	1.4	70.98
06-127	123	256.0	408	458	1.26	3.4	62.10
06-128	125	243.0	360	389	0.97	2.4	61.43
06-129	128	257.0	367	389	0.91	1.0	57.75
06-130	107	242.0	345	388	0.85	.	.
06-131	102	261.0	422	451	1.33	3.8	54.35
06-132	131	210.0	324	357	0.94	1.9	60.02
06-133	114	235.0	348	394	0.93	1.9	51.98

Lab #	Tag #	SOT Wt (kg)	EOT Wt (kg)	Ship Wt (kg)	ADG (kg/day)	BF (mm)	REA (cm^2)
06-134	103	214.0	339	389	1.03	2.4	51.71
06-135	74	248.0	366	390	0.97	1.0	69.76
06-136	147	259.0	413	451	1.27	1.4	71.06
06-137	164	214.0	347	403	1.10	2.4	57.66
06-138	146	258.0	411	464	1.26	3.9	61.15
06-139	162	243.0	364	408	1.00	1.0	59.38
06-140	144	258.0	414	457	1.29	5.3	68.90
06-141	163	248.0	360	392	0.93	3.4	49.47
06-142	148	271.0	397	447	1.04	1.0	60.06
06-143	158	262.0	397	445	1.12	4.3	60.33
06-144	140	241.0	375	429	1.11	2.4	59.50
06-145	145	213.0	368	408	1.28	2.9	56.34
06-146	136	238.0	405	442	1.38	3.4	60.23
06-147	155	215.0	344	391	1.07	1.0	66.61
06-148	142	229.0	342	397	0.93	1.9	59.47
06-149	152	242.0	394	424	1.26	2.5	73.98
06-150	159	256.0	408	457	1.26	2.0	65.69
06-151	138	241.0	400	436	1.31	4.8	63.21
06-152	139	272.0	389	431	0.97	1.9	69.57
06-153	156	251.0	414	459	1.35	3.4	64.20
06-154	135	241.0	368	411	1.05	2.0	63.10
06-155	137	225.0	372	421	1.21	2.4	61.84
06-156	166	249.0	417	468	1.39	1.9	57.15
06-157	165	252.0	371	416	0.98	1.0	63.70
06-158	149	220.0	381	428	1.33	0.0	64.45
06-159	151	235.0	353	400	0.98	3.8	56.63
06-160	143	270.0	444	485	1.44	0.0	63.60
06-161	161	269.0	410	456	1.17	1.9	59.91
06-162	160	241.0	425	475	1.52	2.4	58.22
06-163	141	280.0	421	475	1.17	2.4	69.11
06-164	153	234.0	340	393	0.88	.	.
06-165	134	221.0	340	384	0.98	0.0	52.50
06-166	154	259.0	406	457	1.21	1.0	72.02
06-167	157	261.0	392	428	1.08	0.0	62.11
06-168	150	216.0	350	388	1.11	3.4	62.27
06-169	221	224.0	330	382	0.88	3.8	57.65
06-170	218	288.0	431	477	1.19	4.4	64.39
06-171	202	274.0	409	442	1.13	1.0	64.17
06-172	201	233.0	369	419	1.13	1.0	62.20
06-173	223	234.0	358	401	1.03	2.4	63.94
06-174	203	241.0	366	426	1.04	0.0	59.22
06-175	216	243.0	363	419	1.00	1.9	61.21
06-176	211	225.0	363	401	1.15	2.0	59.38
06-177	210	238.0	377	417	1.16	4.8	66.52
06-178	215	252.0	400	445	1.23	1.4	61.70

Lab #	Tag #	SOT Wt (kg)	EOT Wt (kg)	Ship Wt (kg)	ADG (kg/day)	BF (mm)	REA (cm^2)
06-179	200	238.0	363	404	1.04	1.0	54.41
06-180	209	221.0	336	380	0.96	2.0	52.75
06-181	212	249.0	360	405	0.93	1.0	57.80
06-182	206	236.0	382	415	1.22	2.4	59.88
06-183	225	231.0	363	402	1.10	7.7	54.86
06-184	229	261.0	397	449	1.13	2.5	64.31
06-185	204	254.0	411	459	1.31	0.0	65.19
06-186	222	263.0	396	444	1.11	1.0	67.37
06-187	219	247.0	375	434	1.07	4.0	54.20
06-188	205	254.0	399	459	1.21	0.0	67.47
06-189	213	254.0	369	409	0.96	1.0	69.87
06-190	208	221.0	367	419	1.22	1.6	62.36
06-191	224	230.0	366	422	1.13	1.9	63.81
06-192	226	255.0	430	486	1.46	2.4	56.99
06-193	217	240.0	384	436	1.20	1.0	70.96
06-194	227	272.0	388	421	0.97	1.4	61.35
06-195	207	259.0	385	434	1.05	1.0	58.29
06-196	220	246.0	366	406	1.00	2.9	59.94
06-197	214	231.0	360	413	1.08	1.0	76.77
06-198	228	216.0	336	377	1.00	1.0	68.68
06-199	180	230.0	376	424	1.22	4.8	68.69
06-200	184	261.0	393	415	1.10	2.9	67.47
06-201	188	262.0	439	482	1.48	3.8	64.80
06-202	186	266.0	407	444	1.18	1.4	70.58
06-203	194	258.0	385	406	1.06	3.4	73.18
06-204	179	225.0	351	399	1.05	1.0	53.36
06-205	174	219.0	371	418	1.26	2.9	60.68
06-206	170	229.0	372	410	1.18	1.4	68.31
06-207	185	202.0	336	371	1.12	1.0	56.70
06-208	196	231.0	378	414	1.23	2.9	53.79
06-209	182	218.0	366	412	1.23	1.0	54.89
06-210	171	204.0	358	380	1.27	3.4	58.68
06-211	183	235.0	408	458	1.44	3.4	70.03
06-212	173	246.0	362	398	0.96	2.9	62.49
06-213	181	238.0	399	438	1.34	1.9	62.05
06-214	191	246.0	417	449	1.43	.	.
06-215	187	241.0	400	444	1.33	3.4	56.68
06-216	199	269.0	423	463	1.28	1.0	77.03
06-217	197	245.0	394	435	1.24	2.4	62.39
06-218	193	245.0	420	477	1.46	1.6	77.33
06-219	189	238.0	395	430	1.31	1.6	65.49
06-220	178	208.0	334	371	1.05	1.6	63.78
06-221	168	237.0	390	440	1.26	4.3	63.38
06-223	195	233.0	398	446	1.38	3.8	61.64
06-224	167	228.0	403	439	1.45	1.0	65.23

Lab #	Tag #	SOT Wt (kg)	EOT Wt (kg)	Ship Wt (kg)	ADG (kg/day)	BF (mm)	REA (cm^2)
06-225	175	253.0	395	438	1.18	1.0	83.09
06-226	169	255.0	438	478	1.51	3.8	64.71
06-227	177	248.0	382	424	1.12	1.0	67.89
06-228	192	239.0	370	409	1.09	1.4	63.47
06-229	176	215.0	316	.	0.84	.	.
06-230	198	231.0	396	424	1.38	1.0	70.26
06-231	190	247.0	388	426	1.18	3.8	67.73
06-232	172	230.0	351	397	1.00	1.6	63.14
06-233	237	273.0	447	506	1.51	4.3	54.78
06-234	258	232.0	369	415	1.19	5.8	58.19
06-235	265	260.0	413	458	1.33	3.8	61.71
06-236	249	258.0	439	497	1.57	1.6	66.83
06-237	236	246.0	402	442	1.36	4.4	61.70
06-238	256	254.0	413	468	1.38	1.0	59.47
06-239	238	236.0	389	441	1.33	2.4	66.69
06-240	239	243.0	419	473	1.53	2.9	67.12
06-241	230	264.0	420	452	1.36	3.4	70.46
06-242	232	234.0	400	466	1.44	5.8	66.39
06-243	233	205.0	350	389	1.26	1.9	57.31
06-244	251	271.0	416	467	1.26	5.8	64.19
06-245	259	242.0	409	450	1.45	4.8	79.65
06-246	243	271.0	392	444	1.05	2.9	59.75
06-247	231	256.0	369	412	0.98	2.9	72.90
06-248	252	250.0	396	434	1.27	5.3	62.98
06-249	262	221.0	373	417	1.32	1.0	63.07
06-250	253	230.0	366	402	1.18	1.0	57.19
06-251	246	263.0	392	436	1.12	3.4	69.58
06-252	241	242.0	362	399	1.04	3.9	57.86
06-253	254	224.0	394	439	1.48	2.9	53.08
06-254	257	234.0	373	409	1.21	2.9	54.45
06-255	267	253.0	391	439	1.20	2.9	61.32
06-256	242	260.0	409	465	1.30	8.2	73.32
06-257	247	248.0	354	389	0.92	3.9	68.77
06-258	255	244.0	403	436	1.38	1.0	59.34
06-259	260	221.0	390	443	1.47	3.8	57.00
06-260	234	213.0	365	411	1.32	1.9	62.01
06-261	264	249.0	407	438	1.37	1.4	75.90
06-262	266	260.0	432	489	1.50	3.8	74.27
06-263	235	274.0	397	443	1.07	3.1	77.34
06-264	268	255.0	415	473	1.39	4.3	58.87
06-265	250	241.0	391	426	1.30	2.9	62.07
06-266	334	226.0	340	376	1.00	1.0	74.66
06-267	330	267.0	398	428	1.15	2.4	68.76
06-268	314	210.0	357	415	1.29	2.5	59.24
06-269	333	235.0	384	424	1.31	1.4	78.61

Lab #	Tag #	SOT Wt (kg)	EOT Wt (kg)	Ship Wt (kg)	ADG (kg/day)	BF (mm)	REA (cm^2)
06-270	328	253.0	407	457	1.35	1.4	65.84
06-271	322	248.0	352	402	0.91	1.0	78.29
06-272	307	286.0	421	460	1.18	2.5	74.98
06-273	336	240.0	372	422	1.16	2.4	59.01
06-274	337	247.0	382	417	1.18	2.4	60.86
06-275	339	253.0	394	435	1.24	3.4	73.54
06-276	315	232.0	396	440	1.44	1.4	84.14
06-277	326	257.0	415	455	1.39	1.6	63.95
06-278	309	277.0	397	438	1.05	1.0	69.29
06-279	325	216.0	343	386	1.11	1.0	66.86
06-280	327	248.0	372	405	1.09	1.0	65.05
06-281	317	187.5	310	.	1.07	.	.
06-282	320	240.0	372	422	1.16	1.0	62.71
06-283	319	220.0	377	424	1.38	1.9	57.06
06-284	323	252.0	372	422	1.05	1.4	67.29
06-285	335	271.0	406	453	1.18	3.8	60.32
06-286	312	227.0	377	423	1.32	1.0	61.09
06-287	340	234.0	364	405	1.14	1.0	63.92
06-288	324	265.0	413	451	1.30	2.9	63.34
06-289	329	260.0	414	474	1.35	2.9	77.50
06-290	308	278.0	418	462	1.23	2.9	76.26
06-291	311	281.0	436	488	1.36	1.0	72.24
06-292	313	262.0	417	455	1.36	1.4	77.95
06-293	310	259.0	388	430	1.13	1.0	57.34
06-294	332	247.0	397	435	1.32	2.4	76.56
06-295	331	195.5	346	394	1.32	3.4	64.12
06-296	316	220.0	373	420	1.34	1.0	73.38
06-297	318	254.0	410	451	1.37	2.9	66.70
06-298	338	249.0	360	404	0.97	1.0	57.08
06-299	321	260.0	410	445	1.32	2.4	61.97
06-300	296	260.0	380	421	1.04	2.0	62.04
06-301	301	257.0	403	434	1.27	4.3	52.79
06-302	294	195.5	329	375	1.16	2.4	47.50
06-303	292	252.0	388	427	1.18	2.9	56.37
06-304	289	234.0	374	415	1.22	2.0	59.98
06-305	263	234.0	412	460	1.55	2.4	60.73
06-306	290	243.0	375	431	1.15	1.9	60.11
06-307	277	261.0	393	436	1.15	1.4	66.19
06-308	304	235.0	377	426	1.23	2.5	62.44
06-309	281	246.0	391	436	1.26	2.9	69.09
06-310	293	257.0	432	462	1.52	4.8	60.39
06-311	269	271.0	420	459	1.30	6.3	53.45
06-312	240	189.0	335	377	1.27	3.8	68.41
06-313	288	238.0	385	418	1.28	2.4	60.59
06-314	274	237.0	352	385	1.00	2.0	63.32

Lab #	Tag #	SOT Wt (kg)	EOT Wt (kg)	Ship Wt (kg)	ADG (kg/day)	BF (mm)	REA (cm^2)
06-315	272	244.0	394	439	1.30	2.4	63.25
06-316	271	251.0	409	456	1.37	4.3	66.24
06-317	306	233.0	382	431	1.30	1.9	70.93
06-318	248	245.0	348	389	0.90	1.4	58.36
06-319	287	251.0	412	447	1.40	1.0	67.57
06-320	280	218.0	346	379	1.11	2.4	57.14
06-321	285	237.0	430	494	1.68	3.4	55.54
06-322	283	225.0	408	450	1.59	4.3	71.76
06-323	295	264.0	451	492	1.63	3.8	63.37
06-324	245	275.0	436	488	1.40	4.8	73.10
06-325	278	264.0	381	415	1.02	3.4	64.12
06-326	291	260.0	399	437	1.21	1.0	70.50
06-327	300	228.0	377	408	1.30	2.5	66.68
06-328	270	232.0	389	445	1.37	2.9	63.04
06-329	305	253.0	391	426	1.20	2.0	58.52
06-330	303	272.0	421	469	1.30	1.4	75.23
06-331	273	259.0	379	413	1.04	3.9	60.12
06-332	261	240.0	382	429	1.23	1.9	53.52
06-333	275	219.0	365	422	1.27	.	.
06-334	348	237.0	369	417	1.16	1.0	73.89
06-335	366	230.0	389	407	1.39	2.3	70.78
06-336	365	223.0	372	416	1.31	1.0	67.96
06-337	350	281.0	419	444	1.21	2.9	89.66
06-338	345	252.0	386	430	1.18	3.4	62.25
06-339	359	248.0	381	428	1.17	2.4	72.81
06-340	347	265.0	406	456	1.24	1.9	72.65
06-341	361	227.0	359	406	1.16	1.0	59.19
06-342	346	249.0	366	412	1.03	1.0	72.00
06-343	372	240.0	393	452	1.34	1.0	58.47
06-344	356	259.0	347	385	0.77	0.0	67.11
06-345	362	230.0	357	413	1.11	0.0	74.56
06-346	369	277.0	392	421	1.01	5.3	63.43
06-347	352	219.0	348	395	1.13	1.4	62.76
06-348	374	222.0	382	417	1.40	3.8	62.22
06-349	373	227.0	355	391	1.12	1.4	65.54
06-350	355	259.0	367	413	0.95	1.0	66.08
06-351	349	308.0	437	479	1.13	3.0	80.61
06-352	353	265.0	395	440	1.14	3.8	60.17
06-353	343	261.0	396	440	1.18	1.9	72.90
06-354	360	255.0	403	451	1.30	0.0	75.55
06-355	354	254.0	380	410	1.11	1.4	63.99
06-356	351	294.0	429	479	1.18	1.9	82.75
06-357	367	250.0	391	437	1.24	1.0	78.89
06-358	370	263.0	387	439	1.09	3.8	71.43
06-359	341	259.0	378	419	1.04	1.0	72.52

Lab #	Tag #	SOT Wt (kg)	EOT Wt (kg)	Ship Wt (kg)	ADG (kg/day)	BF (mm)	REA (cm^2)
06-360	358	224.0	347	393	1.08	1.6	50.25
06-361	371	211.0	338	374	1.11	2.9	62.64
06-362	344	237.0	384	437	1.29	2.9	54.69
06-363	342	233.0	349	379	1.02	1.0	53.91
06-364	363	275.0	405	442	1.14	2.9	72.96
06-365	364	247.0	381	410	1.18	2.4	66.21
06-366	357	247.0	414	477	1.46	2.0	67.17
06-367	298	241.0	339	380	0.85	2.0	54.07
06-368	279	239.0	350	395	0.97	1.0	62.77
06-369	385	261.0	405	470	1.26	3.8	66.46
06-370	276	218.0	327	353	0.95	2.9	52.30
06-371	384	246.0	384	426	1.21	1.4	67.56
06-372	388	243.0	377	431	1.18	4.8	56.18
06-373	394	231.0	388	448	1.38	1.0	62.15
06-374	284	244.0	387	439	1.24	1.4	59.70
06-375	380	232.0	356	398	1.09	4.3	61.75
06-376	397	248.0	382	427	1.18	1.0	61.08
06-377	282	257.0	392	412	1.17	2.4	70.90
06-378	378	259.0	388	441	1.13	3.8	65.80
06-379	375	247.0	365	389	1.04	0.0	53.08
06-380	400	239.0	353	403	1.00	2.4	61.41
06-381	379	248.0	383	428	1.18	1.6	67.21
06-382	386	249.0	357	391	0.95	2.9	67.62
06-383	286	283.0	388	407	0.91	3.4	67.10
06-384	391	237.0	359	398	1.07	4.8	63.86
06-385	377	233.0	364	414	1.15	1.0	54.16
06-386	398	272.0	401	437	1.13	1.6	78.50
06-387	390	276.0	414	472	1.21	1.4	82.11
06-388	299	224.0	331	368	0.93	1.0	58.33
06-389	399	242.0	367	397	1.10	1.0	68.96
06-390	382	245.0	394	433	1.31	1.0	63.58
06-391	376	257.0	392	437	1.18	1.0	65.16
06-392	383	248.0	405	446	1.38	2.4	69.54
06-393	297	245.0	371	399	1.10	2.9	58.96
06-394	393	217.0	346	400	1.13	1.0	59.13
06-395	381	231.0	324	362	0.82	1.4	54.18
06-396	389	249.0	360	415	0.97	2.0	68.88
06-397	396	243.0	387	433	1.26	2.4	77.61
06-398	387	233.0	347	402	1.00	1.0	72.84
06-399	302	201.0	293	330	0.80	1.0	57.17
06-400	395	204.0	313	354	0.96	1.0	65.76
06-401	392	216.0	320	368	0.91	2.0	56.64

Appendix F Finishing Growth & Carcass Data

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-001	12	705.34	1.75	377.6	10	8	97	8	AA	64
06-002	7	675.85	1.68	392.6	4	4	115	8	AA	64
06-003	5	594.21	1.48	356.8	5	5	117	9	A	65
06-004	20	700.80	1.72	425.0	8	7	112	8	AA	64
06-005	25	612.35	1.53	360.4	7	5	99	8	AA	64
06-006	19	601.01	1.52	350.4	9	7	86	8	AA	62
06-007	23	573.79	1.24	341.3	12	10	91	8	AA	58
06-008	18	777.91	1.96	460.6	13	11	108	7	AAA	58
06-009	27	.	.	.	.	.	.	.	.	.
06-010	3	698.53	1.96	396.2	8	5	86	7	AAA	64
06-011	21	648.64	1.59	389.9	14	10	114	8	AA	61
06-012	33	696.26	1.87	414.8	14	13	96	8	AA	57
06-013	26	625.96	1.43	370.3	7	5	117	8	AA	64
06-014	4	621.42	1.23	360.2	9	7	99	7	AAA	63
06-015	15	657.71	1.78	400.5	6	5	116	8	AA	64
06-016	17	739.36	1.78	407.6	10	8	96	7	AAA	61
06-017	14	653.17	1.71	380.0	3	2	116	9	A	65
06-018	29	578.33	1.33	369.2	14	13	94	7	AAA	57
06-019	13	684.92	1.63	413.5	10	9	108	8	AA	61
06-020	2	705.34	1.95	415.9	12	11	103	7	AAA	60
06-021	16	.	.	.	.	.	.	.	.	.
06-022	8	648.64	1.62	383.5	7	6	111	7	AAA	62
06-023	1	478.54	0.89	294.8	9	8	99	8	AA	62
06-024	10	594.21	1.49	367.1	12	10	104	8	AA	60
06-025	30	698.53	1.70	423.4	16	14	93	7	AAA	55
06-026	28	659.98	1.63	386.9	8	7	97	7	AAA	62
06-027	32	712.14	1.89	420.7	11	9	108	7	AAA	61
06-028	9	612.35	1.35	381.7	16	15	98	7	AAA	58

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-029	31	671.32	1.62	399.4	8	6	106	7	AAA	62
06-030	24	759.77	2.22	431.6	14	12	95	7	AAA	57
06-031	22	619.15	1.44	378.3	6	4	103	8	AA	64
06-032	11	625.96	1.70	374.2	7	5	104	8	AA	64
06-033	6	616.89	1.62	371.0	5	5	103	8	AA	62
06-034	65	641.83	1.49	372.9	6	5	99	7	AAA	64
06-035	41	623.69	1.81	359.2	14	12	98	8	AAA	59
06-036	39	553.38	1.05	332.0	3	2	98	8	AA	65
06-037	64	675.85	1.72	395.5	8	7	101	7	AAA	62
06-040	50	659.98	1.62	379.0	16	11	98	7	AAA	59
06-041	55	650.91	1.68	370.1	9	8	96	7	AAA	62
06-042	52	657.71	1.80	392.4	8	9	93	7	AAA	59
06-043	54	605.55	1.46	365.8	5	4	99	8	AA	64
06-044	51	621.42	1.92	371.0	18	20	85	7	AAA	51
06-045	48	675.85	1.65	396.7	7	7	97	.	B4	C
06-046	40	573.79	1.00	342.0	5	4	109	8	AA	65
06-047	45	730.28	2.12	424.1	6	6	98	7	AAA	64
06-048	43	771.11	1.92	444.1	7	7	104	8	AA	62
06-049	49	712.14	1.90	420.9	10	9	99	7	AAA	61
06-050	61	657.71	1.77	406.4	8	7	109	8	AA	64
06-051	59	571.53	1.29	357.9	5	4	98	8	AA	63
06-052	38	566.99	1.42	347.0	8	6	103	8	AA	64
06-053	46	562.45	1.42	350.6	6	5	116	8	AA	64
06-054	53	535.24	1.43	320.9	15	14	80	7	AAA	57
06-055	67	553.38	1.19	325.5	9	5	88	8	AA	62
06-056	56	610.08	1.47	376.0	10	7	102	7	AAA	62
06-057	47	705.34	1.85	427.3	11	9	110	8	AA	61
06-058	57	700.80	1.69	389.4	19	20	76	7	AAA	50
06-059	63	585.13	1.36	350.9	3	3	98	9	A	64
06-060	58	691.73	1.93	396.7	14	14	95	8	AA	55
06-061	34	743.89	2.23	442.0	9	7	124	8	AA	62
06-062	42	576.06	1.51	339.3	14	12	90	8	AA	58

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-063	62	743.89	2.00	411.9	12	10	93	7	AAA	60
06-064	35	721.21	2.08	411.6	10	8	115	8	AA	62
06-065	36	732.55	1.80	426.6	5	5	107	8	AA	64
06-066	60	530.70	1.43	318.7	12	8	79	7	AAA	60
06-067	44	669.05	1.89	388.3	7	5	98	8	AA	64
06-068	66	641.83	1.58	387.1	11	8	88	8	AA	60
06-069	37	571.53	1.03	360.2	4	4	97	8	AA	64
06-070	76	784.71	2.21	444.8	15	14	93	.	B4	C
06-071	81	671.32	1.85	398.3	13	12	96	7	AAA	58
06-072	78	616.89	1.76	359.9	10	8	83	7	AAA	62
06-073	97	619.15	1.54	386.9	7	5	109	7	AAA	64
06-074	79	605.55	1.50	364.9	7	5	95	8	AA	64
06-075	86	623.69	1.71	369.2	12	10	91	8	AA	61
06-076	96	582.87	1.36	367.6	5	3	114	8	AA	65
06-077	68	594.21	1.34	362.7	7	6	109	8	AA	64
06-078	87	573.79	1.42	327.5	13	10	91	8	AA	60
06-079	93	571.53	1.45	365.4	5	4	112	7	AAA	64
06-080	69	621.42	1.56	353.8	16	12	102	8	AA	59
06-081	98	635.03	1.59	.	.	.	.	.	.	.
06-082	71	655.44	1.64	377.4	12	9	93	8	AA	60
06-083	99	578.33	1.49	361.5	7	6	113	8	AA	64
06-084	94	571.53	1.32	348.1	11	9	85	7	AAA	62
06-085	90	587.40	1.27	355.8	10	8	105	8	AAA	62
06-086	95	548.85	1.25	332.5	5	4	99	9	A	63
06-087	73	709.87	2.12	420.9	15	16	98	7	AAA	54
06-088	70	557.92	1.40	354.5	6	5	115	8	AA	65
06-089	77	557.92	1.12	329.8	4	2	108	9	A	65
06-090	85	580.60	1.70	353.6	5	4	101	9	A	64
06-091	83	707.60	1.84	420.0	16	13	92	7	AAA	57
06-092	82	725.75	2.10	412.1	7	4	121	8	AA	64
06-093	84	632.76	1.94	383.5	16	12	83	7	AAA	57
06-094	91	632.76	1.58	384.0	7	5	114	8	AA	64

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-095	88	648.64	1.80	398.7	13	12	106	7	AAA	58
06-096	72	.	.	.	.	.	.	.	.	.
06-097	89	752.96	2.11	444.8	11	9	116	8	AA	61
06-098	100	569.26	1.50	344.3	4	3	101	8	AA	65
06-099	92	.	.	.	.	.	.	.	.	.
06-100	80	603.28	1.53	365.6	11	7	107	7	AAA	64
06-101	75	594.21	1.54	374.7	5	4	125	9	A	64
06-102	110	625.96	1.60	386.7	7	4	123	8	AA	62
06-103	109	673.58	1.67	382.2	10	10	93	7	AAA	58
06-104	116	623.69	1.71	350.6	11	10	91	8	AA	59
06-105	118	716.68	1.98	421.4	11	9	117	8	AA	64
06-106	133	616.89	1.47	392.1	5	4	128	8	AA	64
06-107	126	625.96	1.56	362.7	9	8	91	7	AAA	61
06-108	112	732.55	2.31	404.6	12	11	87	8	AA	57
06-109	120	594.21	1.57	361.3	10	8	98	8	AA	62
06-110	124	680.39	1.86	392.8	14	10	89	8	AA	60
06-111	104	689.46	1.70	404.4	11	10	92	7	AAA	60
06-112	113	569.26	1.51	349.0	7	5	117	8	AA	64
06-113	122	635.03	1.88	373.8	11	11	87	8	AA	58
06-114	121	560.19	1.36	365.4	8	6	112	8	AA	64
06-115	117	589.67	1.73	395.2	10	8	86	7	AAA	60
06-116	115	637.30	1.55	372.9	8	7	104	8	AA	64
06-117	129	632.76	1.64	377.2	7	5	116	8	AA	64
06-118	130	610.08	1.50	375.4	8	7	105	8	AA	64
06-119	127	650.91	1.75	383.7	9	7	101	8	AA	64
06-120	132	619.15	1.62	357.0	14	13	96	7	AAA	58
06-121	108	621.42	1.57	379.7	9	7	119	7	AAA	62
06-122	106	589.67	1.56	350.4	5	5	96	8	AA	62
06-123	105	650.91	1.63	385.6	14	12	84	7	AAA	57
06-124	101	551.11	1.44	325.9	11	8	97	8	AA	61
06-125	119	587.40	1.57	346.1	9	8	82	8	AA	59
06-126	111	639.57	1.69	391.2	9	8	107	8	AA	61

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-127	123	675.85	1.76	396.4	8	6	105	8	AA	62
06-128	125	571.53	1.48	348.1	11	9	97	8	AA	60
06-129	128	605.55	1.75	382.4	13	10	89	8	AA	58
06-130	107	560.19	1.39	338.4	7	5	104	9	A	64
06-131	102	669.05	1.77	379.4	14	12	81	8	AA	56
06-132	131	519.36	1.32	318.9	5	4	96	8	AA	64
06-133	114	603.28	1.70	362.7	11	9	93	8	AA	61
06-134	103	582.87	1.57	338.8	13	12	97	8	AA	59
06-135	74	548.85	1.29	.	.	.	.	.	.	.
06-136	147	635.03	1.49	371.7	8	7	110	7	AAA	64
06-137	164	596.47	1.57	353.8	7	8	107	8	AA	62
06-138	146	689.46	1.83	399.8	12	13	101	8	AA	58
06-139	162	598.74	1.55	366.1	9	7	96	7	AAA	62
06-140	144	653.17	1.59	391.5	17	15	103	7	AAA	55
06-141	163	578.33	1.51	352.4	9	8	101	7	AAA	60
06-142	148	687.19	1.95	392.4	9	7	94	7	AAA	61
06-143	158	646.37	1.63	377.6	11	10	88	7	AAA	58
06-144	140	630.49	1.63	376.9	10	8	89	7	AAA	60
06-145	145	598.74	1.55	371.0	9	7	93	7	AAA	64
06-146	136	650.91	1.69	387.4	14	15	87	7	AAA	55
06-147	155	603.28	1.72	378.8	8	6	106	8	AA	64
06-148	142	612.35	1.74	374.4	9	7	106	7	AAA	62
06-149	152	605.55	1.47	360.2	9	7	99	8	AA	61
06-150	159	669.05	1.72	396.1	7	5	96	8	AA	64
06-151	138	657.71	1.80	389.4	12	10	102	7	AAA	59
06-152	139	623.69	1.56	371.3	14	12	88	7	AAA	58
06-153	156	703.07	1.98	406.4	12	10	79	8	AA	56
06-154	135	585.13	1.41	359.7	7	5	109	8	AA	64
06-155	137	616.89	1.59	349.3	6	5	97	8	AA	61
06-156	166	673.58	1.67	397.4	12	9	94	8	AA	61
06-157	165	657.71	1.96	392.4	9	7	115	9	A	64
06-158	149	582.87	1.25	341.6	4	3	108	7	AAA	65

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-159	151	582.87	1.48	366.5	15	15	82	8	AA	54
06-160	143	730.28	1.99	433.2	9	6	107	8	AA	64
06-161	161	657.71	1.63	404.4	6	5	112	8	AA	64
06-162	160	732.55	2.09	433.6	10	9	109	8	AA	64
06-163	141	732.55	2.09	451.1	11	7	114	8	AA	62
06-164	153	591.94	1.61	361.0	8	7	92	8	AA	62
06-165	134	526.17	1.15	325.5	4	3	98	8	AA	64
06-166	154	682.66	1.83	387.6	6	5	118	8	AA	64
06-167	157	635.03	1.68	376.5	5	4	102	8	AA	64
06-168	150	560.19	1.39	341.6	13	12	83	8	AA	57
06-169	221	553.38	1.39	334.8	8	6	84	8	AA	62
06-170	218	725.75	2.02	421.2	19	17	94	7	AAA	54
06-171	202	666.78	1.82	385.6	6	5	104	8	AA	64
06-172	201	564.72	1.18	336.1	2	1	117	.	B4 C	MF
06-173	223	578.33	1.44	365.8	10	7	104	8	AA	62
06-174	203	635.03	1.69	381.0	6	5	107	8	AA	64
06-175	216	580.60	1.31	341.6	5	4	116	8	AA	65
06-176	211	603.28	1.64	358.1	6	4	85	7	AAA	64
06-177	210	551.11	1.09	331.8	13	12	96	8	AA	58
06-178	215	678.12	1.89	398.7	5	4	96	7	AAA	64
06-179	200	616.89	1.72	378.3	10	7	92	8	AA	63
06-180	209	528.44	1.20	320.7	8	7	87	8	AA	62
06-181	212	596.47	1.55	350.2	11	9	94	8	AA	58
06-182	206	621.42	1.67	363.8	9	8	86	8	AA	60
06-183	225	589.67	1.52	360.2	16	17	78	8	AA	53
06-184	229	671.32	1.80	381.9	10	11	84	8	AA	57
06-185	204	721.21	2.12	432.1	8	7	110	8	AA	64
06-186	222	675.85	1.88	411.4	8	6	118	8	AA	64
06-187	219	678.12	1.98	406.0	15	16	91	7	AAA	55
06-188	205	682.66	1.81	387.6	5	4	123	8	AA	64
06-189	213	630.49	1.79	394.6	10	7	106	7	AAA	64
06-190	208	635.03	1.75	388.7	8	6	104	8	AA	62

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-191	224	644.10	1.80	384.0	6	5	99	8	AA	64
06-192	226	741.62	2.07	432.1	10	9	93	7	AAA	60
06-193	217	680.39	1.98	395.1	15	12	93	8	AA	58
06-194	227	578.33	1.27	342.2	7	5	98	8	AA	64
06-195	207	623.69	1.54	373.8	9	6	99	8	AA	63
06-196	220	594.21	1.52	372.9	10	8	113	8	AA	62
06-197	214	603.28	1.54	387.8	5	5	129	9	A	64
06-198	228	657.71	2.27	377.4	8	7	103	7	AAA	62
06-199	180	603.28	1.45	350.2	16	15	89	8	AA	55
06-200	184	614.62	1.62	373.3	10	8	124	7	AAA	64
06-201	188	716.68	1.90	400.1	16	14	91	8	AA	57
06-202	186	632.76	1.53	366.5	6	5	121	8	AA	64
06-203	194	619.15	1.73	395.5	12	10	115	8	AA	61
06-204	179	621.42	1.80	.	.	.	.	.	.	.
06-205	174	591.94	1.41	362.9	3	3	102	.	B4	C
06-206	170	607.81	1.60	377.8	15	14	87	7	AAA	58
06-207	185	594.21	1.81	340.2	12	11	88	7	AAA	58
06-208	196	603.28	1.53	350.6	16	15	85	8	AA	55
06-209	182	605.55	1.57	367.4	13	14	97	7	AAA	57
06-210	171	555.65	1.42	342.5	9	8	101	8	AA	62
06-211	183	646.37	1.53	368.1	7	5	113	7	AAA	64
06-212	173	580.60	1.48	352.0	10	9	91	7	AAA	62
06-213	181	662.24	1.82	389.0	10	8	86	7	AAA	62
06-214	191	689.46	1.95	422.3	7	5	123	8	AA	64
06-215	187	630.49	1.51	364.9	10	8	81	8	AA	59
06-216	199	712.14	2.02	418.9	6	5	128	8	AA	64
06-217	197	625.96	1.55	365.4	7	6	94	8	AA	61
06-218	193	671.32	1.57	397.8	7	5	125	8	AA	64
06-219	189	648.64	1.77	370.6	14	12	98	8	AA	59
06-220	178	551.11	1.46	334.5	6	5	109	8	AA	64
06-221	168	687.19	2.00	393.5	12	11	92	7	AAA	58
06-223	195	610.08	1.33	358.1	11	11	87	8	AA	56

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-224	167	694.00	2.07	408.0	8	7	108	7	AAA	62
06-225	175	630.49	1.56	398.0	3	2	125	9	A	65
06-226	169	666.78	1.53	406.2	12	9	106	7	AAA	62
06-227	177	635.03	1.71	388.7	4	4	115	8	AA	64
06-228	192	621.42	1.72	377.8	6	5	97	8	AA	62
06-229	176	.	.	.	.	.	.	.	.	.
06-230	198	621.42	1.60	367.0	7	5	116	.	B4	C
06-231	190	648.64	1.80	390.8	14	15	104	7	AAA	58
06-232	172	564.72	1.36	345.6	9	7	99	7	AAA	62
06-233	237	737.09	1.87	415.5	20	18	123	7	AAA	54
06-234	258	596.47	1.47	359.0	16	15	83	7	AAA	55
06-235	265	662.24	1.65	375.4	9	8	103	7	AAA	61
06-236	249	728.02	1.87	422.1	9	7	103	7	AAA	64
06-237	236	625.96	1.49	359.8	9	8	91	8	AA	63
06-238	256	666.78	1.61	386.2	10	9	98	8	AA	62
06-239	238	.	.	.	.	.	.	.	.	.
06-240	239	680.39	1.68	401.9	14	12	96	8	AA	58
06-241	230	671.32	1.78	389.9	11	9	102	7	AAA	59
06-242	232	655.89	1.54	388.1	19	19	87	7	AAA	53
06-243	233	566.99	1.44	337.0	7	6	94	8	AA	63
06-244	251	641.83	1.42	373.8	15	13	88	7	AAA	57
06-245	259	680.39	1.87	400.8	13	13	107	7	AAA	58
06-246	243	664.51	1.79	392.2	12	12	93	7	AAA	57
06-247	231	580.60	1.37	359.7	13	12	98	8	AA	58
06-248	252	662.24	1.85	365.4	14	13	96	7	AAA	58
06-249	262	.	.	.	.	.	.	.	.	.
06-250	253	571.53	1.37	339.3	6	5	87	8	AA	64
06-251	246	657.71	1.80	376.9	14	13	99	7	AAA	57
06-252	241	646.37	2.00	368.5	13	13	101	8	AA	58
06-253	254	664.51	1.83	374.7	12	10	79	7	AAA	61
06-254	257	601.01	1.56	358.1	11	9	99	7	AAA	52
06-255	267	607.81	1.37	366.1	9	8	92	8	AA	60

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-256	242	696.26	1.87	408.0	16	14	96	7	AAA	54
06-257	247	523.90	1.09	330.7	11	9	107	.	B4	C
06-258	255	648.64	1.72	402.8	8	5	105	8	AA	62
06-259	260	662.24	1.78	386.5	10	9	97	8	AA	62
06-260	234	612.35	1.63	361.1	9	7	104	8	AA	62
06-261	264	628.23	1.54	386.9	9	8	111	8	AA	62
06-262	266	712.14	1.81	422.5	12	10	114	8	AA	61
06-263	235	689.46	2.00	415.0	14	11	113	7	AAA	59
06-264	268	707.60	1.90	423.0	16	14	96	7	AAA	55
06-265	250	601.01	1.42	353.4	13	8	95	7	AA	60
06-266	334	591.94	1.75	366.7	4	4	121	9	A	65
06-267	330	632.76	1.66	385.8	10	8	114	8	AA	61
06-268	314	630.49	1.75	389.0	8	7	122	8	AA	62
06-269	333	616.89	1.56	397.1	8	7	131	8	AA	64
06-270	328	673.58	1.75	391.9	9	8	103	7	AAA	62
06-271	322	571.53	1.37	370.4	4	3	117	9	A	64
06-272	307	680.39	1.79	424.8	8	5	118	8	AA	64
06-273	336	614.62	1.56	373.5	10	10	104	8	AA	59
06-274	337	582.87	1.34	344.1	9	7	87	7	AAA	61
06-275	339	632.76	1.60	383.5	13	10	99	8	AA	61
06-276	315	680.39	1.95	423.9	7	6	117	.	B4	C
06-277	326	657.71	1.64	396.0	11	8	108	7	AAA	62
06-278	309	659.98	1.80	416.9	9	7	117	8	AA	62
06-279	325	582.87	1.59	350.6	9	7	96	8	AA	64
06-280	327	585.13	1.46	359.2	5	4	128	8	AA	64
06-281	317	.	.	.	.	.	.	.	.	.
06-282	320	673.58	2.04	394.2	13	11	101	8	AA	61
06-283	319	641.83	1.76	377.6	14	12	91	7	AAA	58
06-284	323	623.69	1.63	387.8	7	8	99	8	AA	60
06-285	335	671.32	1.77	386.2	16	16	83	7	AAA	54
06-286	312	589.67	1.35	361.1	10	8	105	8	AA	62
06-287	340	607.81	1.64	383.3	10	9	106	8	AA	61

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-288	324	659.98	1.69	393.7	19	18	95	8	AA	54
06-289	329	678.12	1.65	415.9	9	7	129	8	AA	64
06-290	308	664.51	1.64	394.0	10	8	115	8	AA	61
06-291	311	725.75	1.93	413.2	10	7	117	8	AA	64
06-292	313	666.78	1.72	417.8	8	6	109	8	AA	64
06-293	310	662.24	1.88	383.1	9	7	115	7	AAA	62
06-294	332	657.71	1.80	403.0	11	11	114	8	AA	61
06-295	331	589.67	1.59	372.9	9	8	109	8	AA	64
06-296	316	619.15	1.61	399.8	6	6	127	9	A	64
06-297	318	655.44	1.66	388.3	14	11	97	7	AAA	60
06-298	338	542.04	1.12	317.5	7	4	84	8	AA	63
06-299	321	616.89	1.39	348.8	12	12	83	7	AAA	57
06-300	296	598.74	1.44	373.8	13	11	94	8	AA	58
06-301	301	585.13	1.22	345.1	17	15	87	7	AAA	57
06-302	294	555.65	1.46	328.4	10	7	83	7	AAA	61
06-303	292	610.08	1.48	377.8	14	14	95	8	AA	58
06-304	289	591.94	1.43	347.7	9	8	96	7	AAA	62
06-305	263	682.66	1.80	399.2	13	13	88	8	AA	57
06-306	290	628.23	1.60	373.8	12	11	97	8	AA	59
06-307	277	646.37	1.70	380.1	14	10	94	8	AA	58
06-308	304	580.60	1.25	342.0	8	8	99	8	AA	62
06-309	281	619.15	1.48	376.0	12	10	97	8	AA	59
06-310	293	671.32	1.70	372.9	13	12	83	8	AA	60
06-311	269	684.92	1.83	385.3	15	13	80	7	AAA	54
06-312	240	555.65	1.45	340.7	5	4	98	9	A	64
06-313	288	635.03	1.76	367.4	12	12	97	7	AAA	58
06-314	274	548.85	1.33	335.7	6	4	94	8	AA	64
06-315	272	648.64	1.70	372.2	15	14	91	8	AA	57
06-316	271	671.32	1.74	389.0	16	16	90	7	AAA	56
06-317	306	.	.	.	.	.	.	.	.	.
06-318	248	560.19	1.39	333.4	8	7	104	8	AA	62
06-319	287	646.37	1.62	368.8	6	5	93	7	AAA	64

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-320	280	587.40	1.69	342.2	11	8	107	8	AA	62
06-321	285	762.04	2.17	446.8	16	16	111	7	AAA	56
06-322	283	662.24	1.72	383.3	13	10	105	8	AA	59
06-323	295	741.62	2.02	430.7	14	12	96	7	AAA	59
06-324	245	705.34	1.76	415.3	13	11	110	7	AAA	59
06-325	278	.	.	323.9	7	5	96	8	AA	64
06-326	291	.	.	392.6	6	5	115	7	AAA	64
06-327	300	582.87	1.42	337.9	9	7	96	9	A	64
06-328	270	601.01	1.26	353.6	13	12	89	7	AAA	60
06-329	305	637.30	1.71	364.0	11	8	94	8	AA	62
06-330	303	675.85	1.68	401.0	6	5	116	8	AA	64
06-331	273	555.65	1.16	323.2	13	9	89	8	AA	61
06-332	261	.	.	408.2	14	13	96	7	AAA	57
06-333	275	598.74	1.43	379.0	3	3	118	8	AA	65
06-334	348	621.42	1.66	403.7	4	4	131	8	AA	64
06-335	366	616.89	1.70	384.2	8	7	107	8	AA	64
06-336	365	655.44	1.94	386.5	10	8	104	8	AA	62
06-337	350	646.37	1.64	383.3	10	8	114	7	AAA	64
06-338	345	644.10	1.73	371.0	12	9	91	8	AA	58
06-339	359	630.49	1.64	382.2	12	10	107	8	AA	61
06-340	347	689.46	1.89	419.6	16	14	123	7	AAA	58
06-341	361	641.83	1.91	386.1	9	9	101	8	AA	60
06-342	346	589.67	1.44	365.8	2	2	123	9	A	65
06-343	372	632.76	1.46	380.8	7	7	93	8	AA	64
06-344	356	562.45	1.44	362.9	4	4	119	8	AA	64
06-345	362	594.21	1.47	383.5	4	2	131	9	A	65
06-346	369	612.35	1.55	371.7	12	10	112	7	AAA	61
06-347	352	630.49	1.91	388.7	10	8	96	8	AA	62
06-348	374	612.35	1.58	369.5	12	10	93	8	AA	61
06-349	373	564.72	1.41	348.4	7	5	101	7	AAA	62
06-350	355	637.30	1.82	383.3	10	8	97	8	AA	62
06-351	349	689.46	1.70	401.2	9	8	99	8	AA	62

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-352	353	625.96	1.51	372.6	13	11	107	8	AA	58
06-353	343	591.94	1.23	380.3	6	6	116	8	AA	64
06-354	360	689.46	1.93	418.2	12	9	108	7	AAA	62
06-355	354	603.28	1.57	357.2	12	9	104	8	AA	61
06-356	351	741.62	2.13	462.4	5	4	123	8	AA	64
06-357	367	662.24	1.82	393.5	6	5	128	8	AA	64
06-358	370	635.03	1.59	381.2	11	10	99	8	AA	61
06-359	341	625.96	1.68	392.4	6	5	123	8	AA	65
06-360	358	589.67	1.59	352.4	9	7	103	8	AA	61
06-361	371	546.58	1.40	317.5	5	4	95	9	A	65
06-362	344	621.42	1.49	370.1	9	7	94	7	AAA	62
06-363	342	557.92	1.45	316.4	9	7	97	8	AA	63
06-364	363	621.42	1.45	371.7	17	16	86	7	AAA	57
06-365	364	585.13	1.42	347.0	9	7	107	9	A	60
06-366	357	732.55	2.07	420.9	8	9	110	.	B4	C
06-367	298	585.13	1.66	329.3	10	7	77	8	AA	62
06-368	279	596.47	1.63	380.6	5	5	114	8	AA	65
06-369	385	687.19	1.76	382.6	11	9	107	7	AAA	60
06-370	276	539.77	1.51	317.1	8	6	94	8	AA	62
06-371	384	641.83	1.75	379.9	9	7	104	8	AA	64
06-372	388	632.76	1.63	378.8	16	16	76	7	AAA	52
06-373	394	730.28	2.29	411.4	9	7	109	8	AA	62
06-374	284	637.30	1.61	371.3	12	9	108	8	AA	61
06-375	380	560.19	1.31	329.8	13	11	97	7	AAA	58
06-376	397	605.55	1.45	375.4	4	4	109	8	AA	65
06-377	282	628.23	1.75	365.4	24	23	93	7	AAA	52
06-378	378	687.19	1.99	413.7	15	12	114	8	AA	58
06-379	375	555.65	1.35	306.2	5	4	88	.	B4	C
06-380	400	539.77	1.11	347.5	11	10	102	8	AA	61
06-381	379	664.51	1.92	388.1	12	12	96	8	AA	60
06-382	386	.	.	352.4	12	11	97	8	AA	58
06-383	286	.	.	368.3	10	8	113	8	AA	62

Lab #	Tag #	Live Wt (kg)	ADG (kg/day)	WCW (kg)	Average Fat	Grade Fat	REA (cm^2)	Marbling	Quality Grade	Cutability
06-384	391	553.38	1.26	349.3	5	4	117	8	AA	64
06-385	377	635.03	1.79	388.5	8	7	95	7	AAA	64
06-386	398	655.44	1.77	390.5	12	10	101	7	AAA	59
06-387	390	682.66	1.71	405.5	7	5	122	8	AA	64
06-388	299	542.04	1.41	324.1	6	5	108	8	AA	64
06-389	399	553.38	1.27	339.5	9	7	105	8	AA	64
06-390	382	632.76	1.62	372.6	7	5	104	8	AA	64
06-391	376	666.78	1.86	392.1	8	7	109	8	AA	62
06-392	383	666.78	1.79	394.6	12	10	106	7	AAA	61
06-393	297	623.69	1.82	376.3	13	13	87	7	AAA	56
06-394	393	594.21	1.57	360.2	6	5	106	8	AA	64
06-395	381	553.38	1.55	320.9	10	7	79	7	AAA	61
06-396	389	610.08	1.58	376.5	7	6	101	8	AA	64
06-397	396	669.05	1.91	408.2	9	8	109	8	AA	64
06-398	387	589.67	1.52	365.1	9	9	114	8	AA	62
06-399	302	503.49	1.41	314.1	4	4	112	8	AA	64
06-400	395	553.38	1.62	335.9	3	2	112	8	AA	65
06-401	392	562.45	1.58	338.6	9	8	93	8	AA	62

Appendix G Abstract

Plant & Animal Genome XV, Jan. 13-17, 2007 in San Diego, CA.

Effects Of Corticotropin-Releasing Hormone And Leptin SNPs On Temperament In Beef Cattle.

Kaley A. Pugh[1], Joseph M.Stookey[2], Fiona C. Buchanan[1].

1. Department of Animal Science, University of Saskatchewan, 51 Campus Dr. Saskatoon, SK. S7N 5A8.
2. Department of Large Animal Clinical Sciences, Western College of Veterinary Medicine, 52 Campus Dr. Saskatoon, SK. S7N 5B4.

Temperament in beef cattle can be defined as an animal's response to handling and is a heritable trait of potential economic importance. Animals with calm temperaments have higher average daily gains in feedlots, cause less damage to facilities and cause fewer injuries to employees. We studied previously reported SNPs in two genes for associations with several measurements of temperament in 400 crossbred beef steers. Since temperament is essentially a stress response behavior, we investigated two SNPs in the corticotrophin-releasing hormone gene (*CRH*) (22C>G & 240C>G) as its hormone product is considered to be a stress hormone. The third SNP was in the leptin gene (*LEP*) (C73T), as it has been associated with growth characteristics and the hormone product of *LEP* affects *CRH* expression. A variety of temperament measurements were conducted including a traditional subjective score of individual's response to restraint, as well as several objective measures that quantify individual's response to handling such as flight speed (the time required to cover 2.9 m after release from a scale) and Movement Measurement Device (the amount of movement by an isolated animal). The behavior measurements were taken three times at two month intervals; they were conducted under a strict protocol of minimal interference with the animals in order to remove any effect of handling differences between sessions. The animals' genotypes at the SNPs were then analyzed as main effects, and significant ($p < 0.05$) effects of *CRH* were found with several temperament measures.

Appendix H Poster

Presented at Plant & Animal Genome XV, Jan. 13-17, 2007 in San Diego, CA.

Effects Of Corticotropin-Releasing Hormone And Leptin SNPs On Temperament In Beef Cattle

Kaley A. Pugh[1], Joseph M. Stookey[2], Fiona C. Buchanan[1].

1. Department of Animal Science, University of Saskatchewan, 51 Campus Dr. Saskatoon, SK. S7N 5A8.
2. Department of Large Animal Clinical Sciences, Western College of Veterinary Medicine, 52 Campus Dr. Saskatoon, SK. S7N 5B4.

Introduction

Temperament in beef cattle can be defined as an animal's response to handling and is a heritable trait of potential economic importance.[1,2] Animals with calm temperaments have higher average daily gains in feedlots,[3,4] cause less damage to facilities and cause fewer injuries to employees.[1] We studied previously reported SNPs in two genes for associations with several measurements of temperament in 400 crossbred beef steers. Since temperament is a stress response behavior, we investigated two SNPs in the corticotrophin-releasing hormone gene (*CRH*) (*C22G*[5] & *C240G*[6]) as its hormone product is considered to be a stress hormone, and has an effect on behaviour.[7] The third SNP was in the leptin gene (*LEP*) (*C73T*[8]). It has been associated with growth characteristics[8] and the hormone product of *LEP* affects *CRH* expression.[9]

Materials & Methods

Animals:
- 400 crossbred beef steers
- ~ 8 months old at start of test

Genotyping:
- *LEP* SNP by previously reported PCR-RFLP[8]
- *CRH* SNPs by previously reported PCR-RFLPs[5,6]

Temperament measurements:
- All measurements repeated 3 times, at 2 month intervals
- Subjective Score
  - Animals' response to restraint in a head gate, rated on a 1-5 scale by one observer
- Strain Gauge
  - Measures the amount of force exerted on the head gate by a restrained animal
  - Output is the absolute value of the forces
- Movement Measurement Device[10]
  - Records the amount of movement by an animal isolated on a scale
  - Primary output is the number of peaks
- Exit Time
  - Measures the time it takes for an animal to cover 2.9m upon release from the scale
  - Output is the time in seconds
- For all measurements except Exit Time, a lower value indicates a calmer animal

Statistical Analysis:
- Mixed procedure of SAS software (9.1) [11]
  - SNP genotypes as fixed effects on temperament measurements; single gene effects and two way interactions between *LEP* and each *CRH* SNP

Results

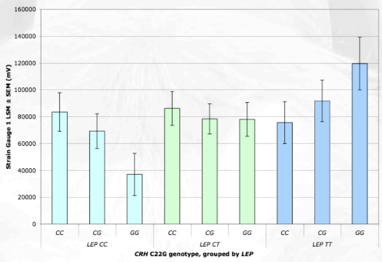

Fig. 1 Interaction between *LEP* and *CRH C22G* for Strain Gauge 1 (P= 0.0398)

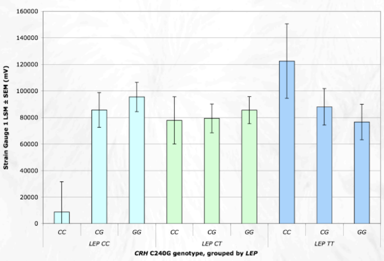

Fig. 2 Interaction between *LEP* and *CRH C240G* for Strain Gauge 1 (P= 0.0158)

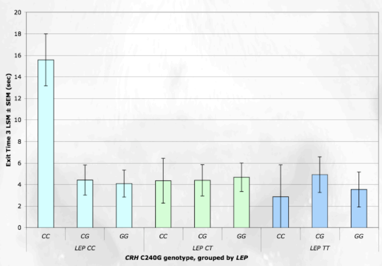

Fig. 3 Interaction between *LEP* and *CRH C240G* for Exit Time 3 (P= 0.0005)

Discussion

- Significant interaction of *LEP* and *CRH C22G* for Strain Gauge 1:
  - For *LEP CC* animals, the *GG* genotype at *CRH C22G* was associated with calmer temperament
  - For *LEP TT* animals, the *CC* genotype at *CRH C22G* was associated with calmer temperament
- Significant interaction of *LEP* and *CRH C240G* for both Strain Gauge 1 and Exit Time 3:
  - For both measurements, the *CC/CC* (*LEP/CRH C240G*) genotype was associated with calmer temperament

Conclusion

- The results from this population of 400 crossbred beef steers indicate that there were significant interactions between the *LEP*, *CRH C22G* and *CRH C240G* SNPs which are associated with the steers' temperaments.
- Calmer animals were:
  - *LEP CC* with *GG* at *CRH C22G*
  - *LEP TT* with *CC* at *CRH C22G*
  - *LEP CC* with *CC* at *CRH C240G*

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